201-14249

To: Rtk Chem/DC/USEPA/US@EPA, Oppt.ncic@epamail.epa.gov
cc: Leslie Scott/DC/USEPA/US@EPA, Richard Hefter/DC/USEPA/US@EPA, Oscar Hernandez/DC/USEPA/US@EPA, santav <santav@cpchem.com>, MARASF <MARASF@cpchem.com>, Craig FARR <craig.farr@atofina.com>, Tom WERKEMA <tom.werkema@atofina.com>, Jim_Keith@americanchemistry.com

Subject: HPV submission - MSA (registration #

Attached is the HPV submission for methane sulfonic acid made on behalf of ATOFINA Chemicals, Inc. and Chevron Phillips LP. Please let me know if you have any problems opening the files.

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MSA-iuclid jan10-2003.zip MSA letter to EPA 1-10-03.zip

ATOFINA

ATOFINA Chemicals, Inc.

January 10, 2002
Submitted electronically to:
Chem.rtk@epa.gov
Oppt.ncic@epa.gov

Christine Todd Whitman, Administrator
U.S. Environmental Protection Agency
P.O. Box 1473
Merrifield, VA 22116

RE: Status of Commitment Under U.S. HPV Program - Registration #
Dear Ms. Whitman:

This submission is being made by ATOFINA Chemicals, Inc and Chevron Phillips Chemical Company LP regarding Methane sulfonic acid (MSA, CAS Number 75-75-2), which is currently sponsored under the U.S. HPV Challenge Program.

Table 1 summarizes the available data for MSA which are described in detail in the attached IUCLID dossier. The data used to characterize the HPV Challenge endpoints for MSA were identified either in company proprietary files, the peer-reviewed literature, and/or calculated using widely accepted computer modeling programs.

Based on the available physical/chemical and human health data, we plan to conduct a 90 day oral toxicity study using OECD Guideline 408 (adopted: 21nd January 1998) which will fulfill the repeated dose toxicity endpoint and provide information on reproductive toxicity. In addition, we will submit a technical discussion for fulfillment of the hydrolysis endpoint.

If you have technical questions, please contact Ann Tveit at (215) 419-5604 or via e-mail at ann.tveit@atofina.com.

Sincerely,

Craig Farr, Ph.D., DABT
Associate Director,
Product Stewardship and Toxicology

Cc: Richard Hefter, EPA

TABLE 1: DATA ANALYSIS/TESTING FOR MSA (CAS# 75-75-2)

CAS No.: 75-75-2	Results	Reliability[1]	Result of Data Review/ Proposed Data Development	References
PHYSICAL/CHEMICAL DATA				
Melting Point	19°C	2	Data Adequate/No testing	Elf Atochem, 1987
Boiling Point	167°C @ 13hPa	2	Data Adequate/No Testing	Merck Index, 1989
Vapor Pressure	0.013 hPa @ 20°C	2	Data Adequate/No Testing	Elf Atochem, 1994
Partition Coefficient (Octanol/Water)	-4.98	2	Data Adequate/No Testing	Leo, 1978
Water Solubility	1000 g/ml	2	Data Adequate/No Testing	ATOFINA, 1987, Lewis. 1993
ENVIRONMENTAL FATE AND PATHWAY				
Aerobic Biodegradability	100% after 28 days	1	Adequate Data / No Testing	Elf Atochem, 1995
Abiotic -Hydrolysis			Technical Discussion Proposed	
Photodegradability	$t_{½}$= 38.75 Days	2	Adequate Data / No Testing	EPIWIN v 3.10
Estimates of Environmental Fate (Fugacity Model – Level III)	Air: 0.4% Water: 45.6% Soil: 54 % Sediment: <0.1%	2	Adequate Data / No Testing	EPIWIN v3.10
ECOTOXICOLOGY DATA:				
Acute Toxicity to Fish	73 mg/l	1	Adequate Data / No Testing	Elf Atochem, 1998
Acute Toxicity to Daphnids	260 mg/l	1	Adequate Data / No Testing	Elf Atochem, 1998
Acute Toxicity to Algae	14	1	Adequate Data / No Testing	Elf Atochem, 199
TOXICOLOGY DATA:				
Acute Toxicity Oral (anhydrous) Dermal (70%)	 LD50= 649 mg/kg LD50 > 1000 mg/kg	 1 1	 Adequate Data / No Testing Adequate Data / No Testing	 Elf Atochem, 1998 Elf Atochem, 2002
Repeated Dose Toxicity OECD 407	NOAEL = 0.026 mg/l	1	Adequate Data / No Testing	Elf Atochem, 1996
Genetic toxicity *Mutagenicity (Ames test)* *Chromosomal Effects (micronucleus)*	 Negative Negative	 1 1	 Adequate Data / No Testing Adequate Data / No Testing	Elf Atochem, 1990 Penwalt, 1989 Penwalt, 1989
Developmental Toxicity OECD 414	NOAEL > 400 mg/kg[2]	1	Adequate Data / No Testing	Elf Atochem, 1996
Reproductive Toxicity	No Data		Testing Proposed (OECD 408)	

[1] All data were evaluated for study reliability in accordance with criteria outlined by USEPA (*Determining the Adequacy of Existing Data*. OPPT, EPA, 1999).

[2] - highest dose tested

I U C L I D

Data Set

Existing Chemical	:	ID: 75-75-2
CAS No.	:	75-75-2
EINECS Name	:	methanesulphonic acid
EC No.	:	200-898-6
Molecular Weight	:	96.1
Structural Formula	:	CH3SO3H
Molecular Formula	:	CH4O3S

Producer related part

Company	:	Atofina
Creation date	:	29.11.2002

Substance related part

Company	:	Atofina
Creation date	:	29.11.2002
Status	:	
Memo	:	
Printing date	:	03.01.2003
Revision date	:	
Date of last update	:	03.01.2003
Number of pages	:	78
Chapter (profile)	:	Chapter: 1, 2, 3, 4, 5
Reliability (profile)	:	Reliability: without reliability, 1, 2, 3, 4
Flags (profile)	:	Flags: without flag, confidential, non confidential, WGK (DE), TA-Luft (DE), Material Safety Dataset, Risk Assessment, Directive 67/548/EEC, SIDS

1.0.1 APPLICANT AND COMPANY INFORMATION

Type	:	cooperating company
Name	:	Atofina
Contact person	:	
Date	:	
Street	:	4-8, cours Michelet La Défense 10
Town	:	95091 Paris La Défense Cedex
Country	:	France
Phone	:	
Telefax	:	
Telex	:	
Cedex	:	
Email	:	
Homepage	:	

27.12.2002

1.0.2 LOCATION OF PRODUCTION SITE, IMPORTER OR FORMULATOR

1.0.3 IDENTITY OF RECIPIENTS

1.0.4 DETAILS ON CATEGORY/TEMPLATE

1.1.0 SUBSTANCE IDENTIFICATION

IUPAC Name	:	Methanesulphonic acid
Smiles Code	:	
Molecular formula	:	C-H4-O3-S
Molecular weight	:	96.11
Petrol class	:	

Source	:	Atofina, Paris-la-Défense, France

27.12.2002

1.1.1 GENERAL SUBSTANCE INFORMATION

Purity type	:	typical for marketed substance
Substance type	:	organic
Physical status	:	liquid
Purity	:	> 99 % w/w
Colour	:	light yellow
Odour	:	Characteristic

Source	:	Atofina, Paris-la-Défense, France
Reliability	:	None of the rats orally exposed to methane sulfonic acid (up to 1800 mg/kg/day) or to its potassium salt (2000 mg/kg/day) died during this 7-day study. Furthermore, none of the measured parameters (food consumption, body weight change, liver and kidney weight) was affected by the exposure. Consequently, NOAEL can be valued at 1 805 mg/kg/day for males and 2122 mg/kg/day for females.

2122 mg/kg/day for females.

27.12.2002

1.1.2 SPECTRA

1.2 SYNONYMS AND TR ADENAMES

methylsulfonic acid, MSA, methanesulfonic acid.

Source : Atofina, Paris-la-Défense, France
27.12.2002

1.3 IMPURITIES

1.4 ADDITIVES

1.5 TOTAL QUANTITY

1.6.1 LABELLING

Labelling	:	as in Directive 67/548/EEC
Specific limits	:	no data
Symbols	:	C, , ,
Nota	:	, D,
R-Phrases	:	(34) Causes burns
S-Phrases	:	(1/2) Keep locked up and out of reach of children (26) In case of contact with eyes, rinse immediately with plenty of water and seek medical advice (36) Wear suitable protective clothing (45) In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible)

Source : Atofina, Paris-la-Défense, France
27.12.2002

1.6.2 CLASSIFICATION

Classified	:	as in Directive 67/548/EEC
Class of danger	:	corrosive
R-Phrases	:	(34) Causes burns
Specific limits	:	
1st Concentration	:	
2nd Concentration	:	
3rd Concentration	:	
4th Concentration	:	
5th Concentration	:	
6th Concentration	:	
7th Concentration	:	
8th Concentration	:	
1st Classification	:	
2nd Classification	:	

3rd	Classification	:
4th	Classification	:
5th	Classification	:
6th	Classification	:
7th	Classification	:
8th	Classification	:

Source : Atofina, Paris-la-Défense, France
27.12.2002

1.6.3 PACKAGING

1.7 USE PATTERN

Type of use : industrial
Category : Basic industry: basic chemicals

27.12.2002

Type of use : industrial
Category : Chemical industry: used in synthesis

27.12.2002

1.7.1 DETAILED USE PATTERN

1.7.2 METHODS OF MANUFACTURE

Origin of substance : Synthesis
Type : Production

Remark : Process description: continuous process; hydrolysis of Methane sulfonyl chloride (130°C); no distillation, stripping of released HCl with water; use of HCl in a other unit on the site.
Source : Atofina, Paris-la-Défense, France
27.12.2002

1.8 REGULATORY MEASURES

1.8.1 OCCUPATIONAL EXPOSURE LIMIT VALUES

1.8.2 ACCEPTABLE RESIDUES LEVELS

1.8.3 WATER POLLUTION

1.8.4 MAJOR ACCIDENT HAZARDS

1.8.5 AIR POLLUTION

1.8.6 LISTINGS E.G. CHEMICAL INVENTORIES

1.9.1 DEGRADATION/TRANSFORMATION PRODUCTS

1.9.2 COMPONENTS

1.10 SOURCE OF EXPOSURE

1.11 ADDITIONAL REMARKS

Memo : Transport information

Remark : UN number: 2586
RID/ADR: class:8
Item (letter): 34°C

Prescriptions: labels: 8.
H.I. Nr/U.N. Nr: 80/2586

IMDG: class: 8
packaging group: III
UN Nr (IMDG): 2586

prescriptions : labels:corrosive/8

IATA: class: 8
packaging group:III
UN Nr: 2586

prescriptions: labels: corrosive/8

Further regulatory information:
RTMD R/F: class: 8
item (letter): 34°C
labels: 8
H.I. Nr/U.N. Nr: 80/2586

Source : Atofina, Paris-la-Défense, France
27.12.2002

1.12 LAST LITERATURE SEARCH

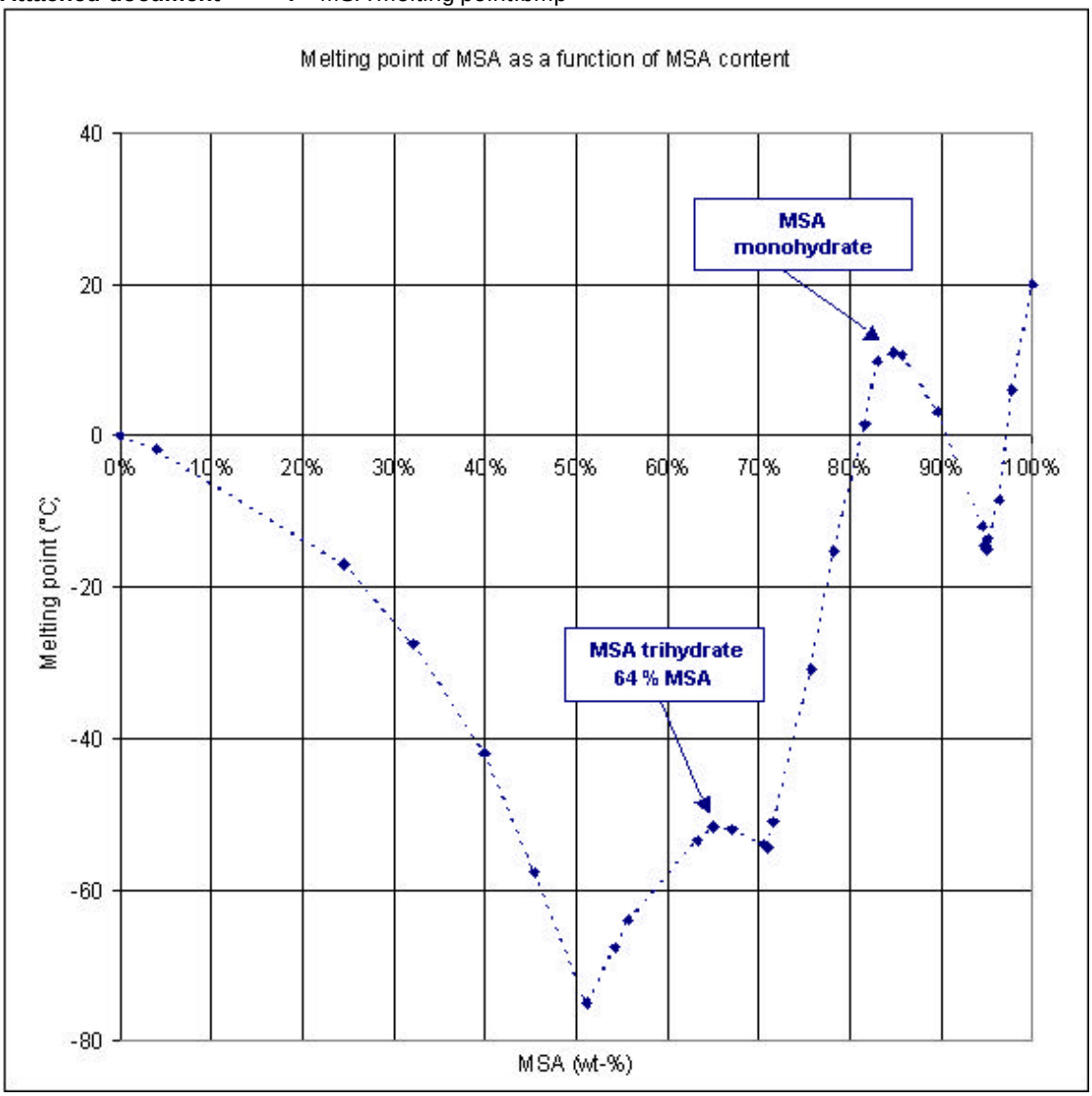
Type of search : Internal and External
Chapters covered : 3, 4, 5
Date of search : 27.12.2002

Source : Atofina, Paris-la-Défense, France
27.12.2002

1.13 REVIEWS

2.1 MELTING POINT

Value	:	= 19 °C
Decomposition	:	yes, at 225 °C
Sublimation	:	
Method	:	other
Year	:	
GLP	:	no data
Test substance	:	other TS
Remark	:	Beginning of decomposition from: 200°C Thermal decomposition giving toxic products: oxides of sulphur. The product is stable at ambient temperature.
Source	:	ATOFINA, Paris-La Défense, France.
Test substance	:	methane sulfonic acid anhydrous
Attached document	:	MSA melting point.bmp



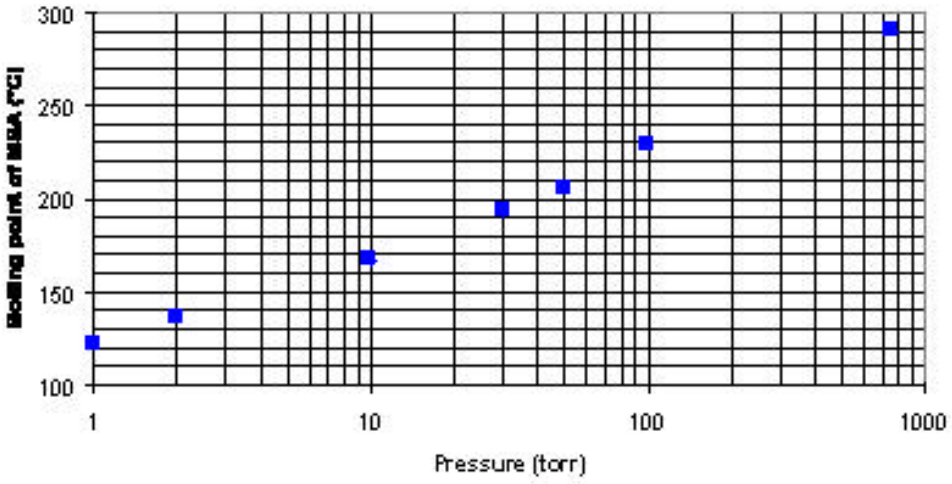
Reliability	:	(2) valid with restrictions
Flag	:	Material Safety Dataset, Critical study for SIDS endpoint

27.12.2002 (1)

2.2 BOILING POINT

Value : 167 °C at 13 hPa
Decomposition : no
Method :
Year :
GLP : no data
Test substance : other TS

Remark : Decomposition 225°C.
Result : 229°C at 130 hPa
Source : ATOFINA, Paris-La Défense, France.
Test substance : Methane sulfonic acid anhydrous.
Attached document : MSA anhydre boiling point.bmp



Reliability : (2) valid with restrictions
data from handbook
Flag : Material Safety Dataset, Critical study for SIDS endpoint
27.12.2002 (2)

Value : = 305 °C at 1033 hPa
Decomposition :
Method : other: calculé
Year :
GLP : no data
Test substance :

Source : ATOFINA, Paris-La Défense, France.
Test substance : Methane sulfonic acid anhydrous.
Reliability : (4) not assignable
accepted calculation method
Flag : Material Safety Dataset
17.12.2002 (3)

2.3 DENSITY

Type : density
Value : = 1481 kg/m3 at 18 °C

Method : other
Year :
GLP : no data
Test substance :

Source : ATOFINA, Paris-La Défense, France.
Test substance : Methane sulfonic acid anhydrous.
Flag : Material Safety Dataset
17.02.1995 (1)

2.3.1 GRANULOMETRY

2.4 VAPOUR PRESSURE

Value : = .013 hPa at 20 °C
Decomposition :
Method : other (measured)
Year :
GLP : no data
Test substance :

Source : ATOFINA, Paris-La Défense, France.
Test substance : Methane sulfonic acid anhydrous.
Flag : Material Safety Dataset, Critical study for SIDS endpoint
17.12.2002 (4)

Value : = 3.97 hPa at 140 °C

Source : ATOFINA, Paris-La Défense, France.
17.12.2002 (5)

2.5 PARTITION COEFFICIENT

Partition coefficient :
Log pow : = -4.98 at °C
pH value :
Method : other (calculated)
Year :
GLP :
Test substance : other TS

Source : ATOFINA, Paris-La Défense, France.
Test substance : Methane sulfonic acid anhydrous
Reliability : (2) valid with restrictions
Accepted calculation method
Flag : Material Safety Dataset, Critical study for SIDS endpoint
27.12.2002 (6)

2.6.1 SOLUBILITY IN DIFFERENT MEDIA

Solubility in : Water
Value : 100 vol% at 20 °C
pH value :
concentration : at °C
Temperature effects :

Examine different pol.	:	
pKa	:	-1.92 at 25 °C
Description	:	miscible
Stable	:	
Source	:	ATOFINA, Paris-La Défense, France.
Test substance	:	Methane sulfonic acid anhydrous
Reliability	:	(2) valid with restrictions
Flag	:	Material Safety Dataset, Critical study for SIDS endpoint

27.12.2002 (7) (8) (9)

Solubility in	:	other: benzene
Value	:	1.5 vol% at 25 °C
pH value	:	
concentration	:	at °C
Temperature effects	:	
Examine different pol.	:	
pKa	:	at 25 °C
Description	:	
Stable	:	
Test substance	:	Methane sulfonic acid anhydrous. Data from handbook

17.12.2002 (2)

2.6.2 SURFACE TENSION

2.7 FLASH POINT

Value	:	= 189 °C
Type	:	closed cup
Method	:	other
Year	:	
GLP	:	no data
Test substance	:	
Source	:	ELF ATOCHEM Paris la defense 10 EUROPEAN COMMISSION - European Chemicals Bureau Ispra (VA)
Test substance	:	Methane sulfonic acid anhydrous.

17.02.1995 (10)

2.8 AUTO FLAMMABILITY

Value	:	> 500 °C at 1013
Method	:	other
Year	:	
GLP	:	no data
Test substance	:	
Source	:	ELF ATOCHEM Paris la defense 10 EUROPEAN COMMISSION - European Chemicals Bureau Ispra (VA)
Test substance	:	Methane sulfonic acid anhydrous.

17.02.1995 (10)

2.9 FLAMMABILITY

2.10 EXPLOSIVE PROPERTIES

2.11 OXIDIZING PROPERTIES

2.12 DISSOCIATION CONSTANT

Acid-base constant : -1.86
Method :
Year :
GLP :
Test substance : other TS

Test substance : Methane sulfonic acid anhydrous.
Reliability : (4) not assignable
27.12.2002 (11)

2.13 VISCOSITY

Value : - 13.5 mPa s (dynamic) at 20 °C
Result :

17.12.2002

2.14 ADDITIONAL REMARKS

3.1.1 PHOTODEGRADATION

Type	:	air
Light source	:	
Light spectrum	:	nm
Relative intensity	:	based on intensity of sunlight
Deg. product	:	
Method	:	other (calculated)
Year	:	
GLP	:	
Test substance	:	

Remark :

AOP Program (v1.90) Results:
==========
SMILES : O=S(=O)(O)C
CHEM : Methanesulfonic acid
MOL FOR: C1 H4 O3 S1
MOL WT : 96.10
------SUMMARY (AOP v1.90): HYDROXYL RADICALS--------
**Hydrogen Abstraction = 0.1360 E-12 cm3/molecule-sec
Reaction with N, S and -OH = 0.1400 E-12 cm3/molecule-sec
Addition to Triple Bonds = 0.0000 E-12 cm3/molecule-sec
Addition to Olefinic Bonds = 0.0000 E-12 cm3/molecule-sec
Addition to Aromatic Rings = 0.0000 E-12 cm3/molecule-sec
Addition to Fused Rings = 0.0000 E-12 cm3/molecule-sec

OVERALL OH Rate Constant = 0.2760 E-12 cm3/molecule-sec
HALF-LIFE = 38.754 Days (12-hr day; 1.5E6 OH/cm3)
..... ** Designates Estimation(s) Using ASSUMED Value(s)
----SUMMARY (AOP v1.90): OZONE REACTION----------

****** NO OZONE REACTION ESTIMATION ******
(ONLY Olefins and Acetylenes are Estimated)

Experimental Database: NO Structure Matches

Reliability	:	(2) valid with restrictions Accepted calculation method
Flag	:	Critical study for SIDS endpoint

03.01.2003

3.1.2 STABILITY IN WATER

3.1.3 STABILITY IN SOIL

3.2.1 MONITORING DATA

3.2.2 FIELD STUDIES

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type	:	fugacity model level III

Media :
Air : % (Fugacity Model Level I)
Water : % (Fugacity Model Level I)
Soil : % (Fugacity Model Level I)
Biota : % (Fugacity Model Level II/III)
Soil : % (Fugacity Model Level II/III)
Method :
Year :

Remark : Level III Fugacity Model (Full-Output):

==========

Chem Name : Methanesulfonic acid
Molecular Wt: 96.1
Henry's LC : 1.26e-008 atm-m3/mole (Henrywin program)
Vapor Press : 0.0267 mm Hg (Mpbpwin program)
Liquid VP : 0.0272 mm Hg (super-cooled)
Melting Pt : 25.8 deg C (Mpbpwin program)
Log Kow : -2.38 (Kowwin program)
Soil Koc : 0.00171 (calc by model)

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	0.394	930	1000
Water	45.6	360	1000
Soil	54	360	1000
Sediment	0.0759	1.44e+003	0

	Fugacity (atm)	Reaction (kg/hr)	Advection (kg/hr)	Reaction (percent)	Advection (percent)
Air	1.25e-011	3.65	49	0.122	1.63
Water	3.71e-013	1.09e+003	566	36.3	18.9
Soil	1.63e-011	1.29e+003	0	43	0
Sediment	3.09e-013	0.454	0.0189	0.0151	0.000629

Persistence Time: 414 hr
Reaction Time: 521 hr
Advection Time: 2.02e+003 hr
Percent Reacted: 79.5
Percent Advected: 20.5

Half-Lives (hr), (based upon Biowin (Ultimate) and Aopwin):
Air: 930
Water: 360
Soil: 360
Sediment: 1440
Biowin estimate: 3.179 (weeks)

Advection Times (hr):
Air: 100
Water: 1000
Sediment: 5e+004

Reliability : (2) valid with restrictions
Accepted calculation method
Flag : Critical study for SIDS endpoint
03.01.2003

3.3.2 DISTRIBUTION

3.4 MODE OF DEGRADATION IN ACTUAL USE

3.5 BIODEGRADATION

Type	:	aerobic
Inoculum	:	domestic sewage
Concentration	:	11.3 mg/l related to COD (Chemical Oxygen Demand) related to
Contact time	:	28 day(s)
Degradation	:	ca. 100 (±) % after 28 day(s)
Result	:	readily biodegradable
Kinetic of testsubst.	:	3 day(s) = 4 % 10 day(s) = 2 % 17 day(s) = 15 % 21 day(s) = 100 % 28 day(s) = 100 %
Control substance	:	Benzoic acid, sodium salt
Kinetic	:	3 day(s) = 97 % 24 day(s) = 100 %
Deg. product	:	not measured
Method	:	OECD Guide-line 301 A (new version) "Ready Biodegradability: DOC Die Away Test"
Year	:	1992
GLP	:	yes
Test substance	:	other TS
Remark	:	Study peer reviewed
Result	:	See attached document LATENT PERIOD: 10 days VALIDITY CRITERIA: - Extreme values difference: <20% - Degradatio n of reference substance in 14 days: >60% - Inhibition: No (Degradation in inhibition flask in 14 days: >25%)
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	INOCULUM/TEST ORGANISM: - Type of sludge: Secondary effluent from a biologic treatment plant. - Sampling site: Versailles. - Preparation of inoculum: The effluent was taken the day before the sowing of flasks and was centrifugated at 20°C during 20 min at 4000g.The residue was diluted with test medium so that the concentration factor in comparison with the taken sample is about 140. - Initial cell concentration: 1.1 10E4 bact/ml. TEST SYSTEM: - Culturing apparatus: 500 ml Erlenmeyer flasks, corked with carded cotton and gauze. Those flasks have been previously sterilised in a oven at 170°C for 1 hour. - Number of culture flasks per concentration: 2. - Aeration device: no data. - Measuring equipment: Jenway 3410 (oxymeter, conductimeter, pH-meter), Maïhak Tocor 100 (DOC analyzer). - Closed vessels used: Yes. INITIAL TEST SUBSTANCE CONCENTR ATION: 11.3 mg DOC / L. METHOD OF PREPARATION OF TEST SOLUTION: A parent solution of methane sulfonic acid (titrated at 94 mg DOC/L) was diluted on the basis of 30 ml in 250 ml medium

diluted on the basis of 30 ml in 250 ml medium

DURATION OF THE TEST: 28 days.

ANALYTICAL PARAMETER: Dissolved Organi c Carbon (DOC) is measured at each sampling time.
DOC degradation percentage is then calculated according the following formula:
Dt = [1- (Ct-Cbt)/(C0-Cb0)]x100
with Ct: mean DOC concentration in culture medium containing test substance at t time
C0:initial mean DOC concentration in culture medium containing test substance
Cbt: mean DOC concentration in the blank culture medium at t time
Cb0: initial mean DOC concentration in the blank culture medium

SAMPLING: 0, 3, 7, 10, 14, 17, 21, 24 and 28 days.

TEST CONDITIONS
- Composition of medium:

.10 ml of solution a:
8.5g KH2PO4
21.75g K2HPO4
33.4 g Na2HPO4,H2O
0.5g NH4Cl
q.s.p 1000ml ultrapure water.

.1 ml of solution b:
27.5g CaCl2 or 36.40g CaCl2,2H2O
in 1000 ml of ultrapure water.

.1 ml of solution c:
22.5g MgSO4,7H2O
in 1000 ml of ultrapure water.

.1 ml of solution d:
0.25g FeCl3,6H2O
in 1000 ml of ultrapure water (this solution was prepared at the beginning of the test).

.ultrapure water q.s.p 1000 ml.

- pH: 7.4
- Additional substrate: No.
- Test temperature: 22+/-2°C.

INTERMEDIATES / DEGRADATION PRODUCTS: Not identified.

NITRATE/NITRITE MEASUREMENT: No.

Test substance : Source: Elf Atochem
Batch number: 2690
Purity: 70%

Attached document : 75-75-2 Biodeg 2765-95-A.bmp

Substance étudiée :	ACIDE METHANE SULFONIQUE
Substance de référence :	Benzoate de sodium

Dosage du COD (mg/l)										
SERIE	JOUR	0	3	7	10	14	17	21	24	28
Blanc	Fb1	0,1	0,1	0,3	0,6	0,8	0,9	0,5	0,5	1,1
	Fb2	0,1	0,1	0,4	0,5	0,7	0,6	0,6	0,4	0,5
	Moyenne Fb	**0,1**	**0,1**	**0,3**	**0,5**	**0,8**	**0,8**	**0,6**	**0,4**	**0,8**
Substance	Ft1	11,5	11,4	11,8	12,0	11,8	10,0	0,7	0,5	0,3
	Ft2	11,9	11,3	11,5	11,9	11,6	11,2	0,5	0,4	0,2
	Ft3									
	Moyenne Ft	**11,7**	**11,3**	**11,6**	**12,0**	**11,7**	**10,6**	**0,6**	**0,4**	**0,2**
Contrôle	Fc	11,5	0,4	0,8	1,0	1,3	0,9	0,9	0,5	0,9
Stérile	Fs	11,7	11,7	12,0	12,6	12,1	13,0	11,4	11,3	11,7
Inhibition	Fi	22,3	12,3	12,9	12,4	12,1	10,2	0,7	0,8	0,4
Adsorption	Fa	11,5	11,8	12,3	12,7	12,4	18,3	11,2	11,1	12,1

Biodégradation Dt (%)										
SERIE	JOUR	0	3	7	10	14	17	21	24	28
Substance	Ft1	0	1	0	-1	3	19	99	99	105
	Ft2	0,	6	6	4	9	12	101	100	105
	Ft3	#N/A	#N/A	#N/A	#N/A	#N/A	#N/A	#N/A	#N/A	#N/A
	Moyenne Ft	0,	4	3	2	6	15	100	100	105
Contrôle	Fc	0,	97	96	96	96	99	97	100	99
Stérile	Fs	0,	-1	-1	-5	2	-6	7	6	5
Inhibition	Fi	0,	45	43	47	49	57	99	98	102
Adsorption	Fa	0,	-3	-5	-7	-2	-10	7	6	1

Conclusion : Aerobic biodegradation study of methane sulfonic acid shows a maximum biodegradation rate in 28 days (100%).
This rate is reached as early as 21 days.

No physico-chemical degradation was observed (no abiotic degradation).

According to the OECD 301 guideline, methane sulfonic acid can therefore be considered as readily biodegradable.

Reliability : (1) valid without restriction
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

27.12.2002 (12)

Type : aerobic
Inoculum : activated sludge, domestic
Contact time : 68 day(s)
Degradation : < 86.6 (±12) % after 55 day(s)
Result :
Kinetic of testsubst. : 30 day(s) < 0 %
36 day(s) < 15 %
40 day(s) < 10 %
50 day(s) < 10 %
53 day(s) ca. 42 %
Control substance : other: no data
Kinetic : %
%
Deg. product : no
Method : OECD Guide-line 303 A "Simulation Test - Aerobic Sewage Treatment: Coupled Unit Test"
Year : 1981
GLP : no data
Test substance : other TS

Remark : Study peer reviewed
Result : See second and third attached document.

Methane sulfonic acid was introduced on day 13. Approximately 40 days were actually required to reach a steady biodeg radation. The first 20 days

were actually required to reach a steady biodeg radation. The first 20 days even showed a slight inhibition.

Source : Atofina, Paris-la-Défense, France

Test condition : INOCULUM/TEST ORGANISM:
- Type of sludge: activated sludge from treatment plant
- Sampling site: Versailles
- Preparation of inoculum: a sixth of the reactor volume is renewed every day
- Initial cell concentration: no data

TEST SYSTEM:
- Culturing apparatus: waste water treatment model (see the first attached document)
- Number of culture flasks per concentration: one
- Aeration device: yes (see the first attached document)
- Measuring equipment: Dohrmann DC 80 analyzer
- Closed vessels used: no

INITIAL TEST SUBSTANCE CONCENTRATION: 20 mg/l

METHOD OF PREPARATION OF TEST SOLUTION:
16 g of methane sulfonic acid (MSA) is dissolved in 1 litre ultrapure water. This solution is then diluted to the tenth in order to reach a 20 mg of organic carbon per litre.

DURATION OF THE TEST: 46 days

ANALYTICAL PARAMETER: COD, %COD (= (COD2-COD1)/COD2 x 100), DR (= [T - (E - E0)]/T x 100
COD2: COD measured at the exit of the model on D-day
COD1: COD measured at the entrance of the model on day D -1
DR: degradation
T: methane sulfonic acid concentration in synthetic waste water (in mg/l)
E: COD at the exit of the test model
E0: COD at the exit of the control model

SAMPLING: From day 0 to day 68

TEST CONDITIONS
- Composition of medium:
Nutritive solution:
. pancreatic pepton (Prolabo): 16 g
. beef meat extract (Biomérieux): 11 g
. urea (Labosi): 3 g
. NaCl: 0.7 g
. CaCl2, 2 H2O: 0.4 g
. MgSO4, 2 H2O: 0.2 g
. ultrapure water: qsp 1 L
Potassium phosphate solution :
. K2HPO4: 2.8 g
. Ultrpure water: qsp 1L
Eventual medium is composed of 10 ml of these two previous solutions, completed with 2 liters of urban water.
- pH: 6.07 to 8.41
- Additional substrate: no data
- Test temperature: 4°C

INTERMEDIATES / DEGRADATION PRODUCTS: Not identified.

NITRATE/NITRITE MEASUREMENT: yes

REFERENCE SUBSTANCE: no data

Test substance : Source: Atochem
Batch number: 89 8015
Purity: no data

Attached document : 75-75-2 Biodeg (23567) Résultats 1.bmp
75-75-2 Biodeg (23567) Résultats 2.bmp

	UNITE 6 (témoin)				UNITE 2 (essai)			
	COD entrée	COD sortie	%COD élimination		COD entrée	COD sortie	%COD élimination	
JOUR								DR
0	105,7				103,0			
1	107,6	25,5	75,9		112,1	28,1	72,7	
2	107,6				112,1			
3	107,6				112,1			
4	111,5	13,3	87,6		110,6	13,5	88,0	
5	111,5	14,8	86,7		110,6	14,5	86,9	
6	109,5	13,7	87,7		105,8	13,3	88,0	
7	109,5	13,9	87,3		105,8	14,4	86,4	
8	104,4	14,2	87,0		111,7	15,4	85,4	
9	104,4				111,7			
10	104,4				111,7			
11	104,9	13,3	87,3		109,5	13,0	88,4	
12	104,9	10,1	90,4		109,5	10,2	90,7	
13	108,1	9,5	90,9	(20 ppm)	140,0	9,5	91,3	
14	108,1	10,0	90,7		140,0	24,8	82,3	26,0
15	112,6	9,8	90,9		136,8	33,1	76,4	-16,5
16	112,6				136,8			
17	112,6				136,8			
18	106,3	9,2	91,8		130,8	31,2	77,2	-10,0
19	106,3	9,5	91,1		130,8	30,7	76,5	-6,0
20	104,8	9,6	91,0		134,8	33,1	74,7	-17,5
21	104,8	9,4	91,0		134,8	34,3	74,6	-24,5
22	109,0	9,3	91,1		137,0	32,2	76,1	-14,5
23	109,0				137,0			
24	109,0				137,0			
25	105,0	8,8	91,9		129,0	32,3	76,4	-17,5
26	105,0	10,6	89,9		129,0	30,4	76,4	1,0
27	105,5	7,5	92,9		134,9	27,3	78,8	1,0
28	105,5	7,6	92,8		134,9	29,5	78,1	-9,5
29	99,3	7,8	92,6		127,8	30,4	77,5	-13,0
30	99,3				127,8			
31	99,3				127,8			
32	99,5	7,3	92,6		122,4	29,5	76,9	-11,0

	UNITE 6 (témoin)			UNITE 2 (essai)			
	COD entrée	COD sortie	%COD élimination	COD entrée	COD sortie	%COD élimination	
JOUR							DR
33	99,5	8,6	91,4	122,4	29,5	75,9	-4,5
34	110,5	8,7	91,3	128,0	28,4	76,8	1,5
35	110,5	8,6	92,2	128,0	26,3	79,5	11,5
36	109,3	8,6	92,2	121,8	29,1	77,3	-2,5
37	109,3			121,8			
38	109,3			121,8			
39	106,7	8,3	92,4	122,1	27,7	77,3	3,0
40	106,7	9,9	90,7	122,1	29,4	75,9	2,5
41	108,0	10,2	90,4	119,3	29,7	75,7	2,5
42	111,0	7,2	93,3	128,0	26,2	78,0	5,0
43	111,0			128,0			
44	114,6	6,8	93,9	130,0	25,4	80,2	7,0
45	114,6			130,0	32,7	74,8	-63,5
46	103,5	7,9	93,1	122,6			
47	103,5	7,3	92,9	122,6	27,1	77,9	1,0
48	106,3	7,3	92,9	122,6	26,5	78,4	4,0
49	106,3	7,3	93,1	122,6	28,5	76,8	4,0
50	116,0	6,5	93,9	136,0	26,7	78,2	-10,0
51	116,0			136,0			
52	116,0			136,0			
53	116,0			136,0			
54	84,9	7,0	94,0	106,7	18,9	86,1	40,3
55	84,9	7,8	90,8	106,7	8,8	91,8	95,0
56	85,0	7,8	90,8	103,0	9,0	91,6	94,0
57	85,0	7,2	91,5	103,0	8,0	92,2	96,0
58	109,4	6,1	92,8	131,7	9,8	90,5	81,5
59	109,4			131,7			
60	109,4			131,7			
61	109,8	7,4	93,2	127,1	12,0	90,9	77,0
62	109,8	8,0	92,7	127,1	17,1	86,5	54,5
63	119,0	8,2	92,5	139,0	11,0	91,3	86,0
64	119,0	8,0	93,3	139,0	11,2	91,9	84,0
65	120,2	10,1	91,5	137,0	13,5	90,3	83,0
66	120,2			137,0			
67	120,2			137,0			
68		9,5	92,1		13,1	90,4	82,0

Conclusion : Mean biodegradation percentage from day 55 to day 68 was 86.6% (sd: 12.0%).
This degradation was obtained at 20 mg of carbon per litre, that is to say 160 mg/l methane sulfonic acid.

Methane sulfonic acid presents a good biodegradation potential in a waste water treatment plant, but only if it arrives steadily at the plant. Actually, sludge adaptation time was found to be very long here. This means ponctual rejects would probably not be treated.

However, test concentration here is rather high, because of the sensitivity of the analysis method. Lower concentrations could have yielded better outcomes.

Reliability : (2) valid with restrictions
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

03.01.2003 (13)

Type	:	aerobic
Inoculum	:	
Deg. product	:	yes
Method	:	other: EPA, Methods for chemical analysis and wastes
Year	:	1979
GLP	:	no data
Test substance	:	other TS
Remark	:	Study peer reviewed
Result	:	COD is not very representative of the potential oxygen demand for MSA. This could possibly due to the oxidation state of the compound or perhaps MSA is not completely oxidized b the potassium dichromate. Biodegradation inhibition was observed at 100 ppm.
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	INOCULUM/TEST ORGANISM: - Type of sludge: no data - Sampling site: Wyandott plant - Preparation of inoculum: no data - Initial cell concentration: no data TEST SYSTEM: - Culturing apparatus: no data. - Number of culture flasks per concentration: no data - Aeration device: no data - Measuring equipment: Ionics TOC analyzer - Closed vessels used: no data INITIAL TEST SUBSTANCE CONCENTRATION: 1, 10, 50 and 100 ppm METHOD OF PREPARATION OF TEST SOLUTION: Methane sulfonic acid (MSA) solutions were prepared by dilution of anhydrous MSA in lab distilled water. DURATION OF THE TEST: 4 weeks. ANALYTICAL PARAMETER: COD, BOD and TOC SAMPLING: Day 0, day 7, day 14 and day 21 TEST CONDITIONS - Composition of medium: no data - pH: no data - Additional substrate: no data - Test temperature: no data INTERMEDIATES / DEGRADATION PRODUCTS: Not identified. NITRATE/NITRITE MEASUREMENT: no data. REFERENCE SUBSTANCE: no data
Test substance	:	Source: no data Batch number: PF 2690 Purity: no data
Conclusion	:	The lack of information does not allow to have a good appraisal of biodegradation. Readers should interpret the results with circumspection since test condition are not detailed enough. This study is mentioned for the record only.
Reliability	:	(4) not assignable Scarce data do not allow a good interpretation of results.

Type	:	aerobic
Inoculum	:	activated sludge, industrial
Contact time	:	20 day(s)
Degradation	:	(±) % after
Result	:	readily biodegradable
Control substance	:	other: none
Kinetic	:	% %
Deg. product	:	not measured
Method	:	other: NEN 3235-5.4 and 3235-5.3
Year	:	
GLP	:	no data
Test substance	:	other TS
Remark	:	Study peer reviewed
Result	:	Discrepancies have been noted between text and table results. Since results presented in the table are more detailed, they are presented in attached document. However, they should be considered with caution. Nevertheless, all results agree with no toxicity of methane sulfonic acid towards inoculum. Inasmuch methane sulfonic acid is nitrogen-free, nitrification is not likely to occur. Anyway, it has been checked.
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	INOCULUM/TEST ORGANISM: - Type of sludge: active sludge from an oxidation ditch. The oxidation ditch is used to treat domestic sewage. - Sampling site: on the premises of TNO (Delft, Netherlands). - Preparation of inoculum: The original sludge (2 g of solid substance /litre) was allowed to settle for 5 minutes and 2 ml of the supernatant was used to inoculate on litre of BOD dilution water. - Initial cell concentration: no data TEST SYSTEM: - Culturing apparatus: BOD bottles - Number of culture flasks per concentration: 4 - Aeration device: no data. - Measuring equipment: oxygen electrode (OXI 191) - Closed vessels used: no data INITIAL TEST SUBSTANCE CONCENTRATION: 4, 8 and 16 mg/l. METHOD OF PREPARATION OF TEST SOLUTION: A stock solution containing 1600 mg methane sulfonic acid per litre BOD dilution water was prepared. DURATION OF THE TEST: 20 days. ANALYTICAL PARAMETER: Dissolved O2 in the medium is measured after 5 and 20 days of exposure. SAMPLING: 0, 5 and 20 days. TEST CONDITIONS - Composition of medium: no data - pH: 7.13 - Additional substrate: . solution containing glucose (3 g/l) and glutamic acid (3 g/l) up to 1 ml/l of the 16 mg/l MSA solution . allylthiourea (1 mg/l added to a 8 mg/l MSA solution in order to detect

. allylthiourea (1 mg/l added to a 8 mg/l MSA solution in order to detect nitrification influence)
- Test temperature: 20C.

INTERMEDIATES / DEGRADATION PRODUCTS: Not identified.

NITRATE/NITRITE MEASUREMENT: yes

REFERENCE SUBSTANCE: none

Test substance : Source: no data
Batch number: no data
Purity: no data
Other: Methane sulfonic acid 70% solution

Attached document : 75-75-2 BOD (TNO).bmp

Table 1 Oxygen concentrations in BOD-bottles determined after 0, 5 and 20 days of incubation with different concentrations of MSA, and the BOD-values calculated. Controls on inoculum activity, toxicity and nitrification are included. Standard deviations in brackets.

Conc. MSA (70%) ($mg.l^{-1}$)	Additions	Oxygen conc. ($mg.l^{-1}$)			BOD ($mg\ O_2.l^{-1}$)		BOD* $mg\ O_2.mg(test\text{-}substance)^{-1}$	
		0 day	5 days	20 days	5 days	20 days	5 days	20 days
0	-	9.1 (0.0)	8.35 (0.06)	5.93 (0.09)	0.75	3.18	-	-
0	gluc+glut	9.1 (0.0)	3.55 (0.06)	0.23 (0.25)	5.55	8.88	-	-
4	-	9.1 (0.0)	8.3 (0.05)	4.93 (0.19)	0.83	4.43	0.03	0.45
8	-	9.1 (0)	8.3 (0.08)	2.9 (0.0)	0.8	6.2	-	0.54
8	ATU**	9.1 (0.0)	8.75 (0.05)	2.93 (0.61)	0.35	6.2	-	0.54
16	-	9.1 (0.0)	8.23 (0.05)	0.45 (0.30)	0.88	8.65	0.1	0.49
16	gluc+glut	9.1 (0)	3.8 (0)	0.25 (0.3)	5.3	8.85	-	-

* The BOD of the control has been substracted.

** Allylthiourea

Conclusion : No significant BOD5 (20°C) was found.
BOD20 (20°C) was found to be 0.51 ± 0.04 mg O2 per mg of methane sulfonic acid.
Methane sulfonic acid could therefore be characterized as readily biodegradable.

Reliability : (3) invalid
Descrepancies have been noticed between text and table results.

03.01.2003 (15)

Type :
Inoculum : activated sludge
Deg. product :

Method	:	other:OECD 301 Modified OECD screening test (1993)
Year	:	1993
GLP	:	
Test substance	:	

Method :

A glass continuous culture (CC) bioreactor was used (see attached document figure 1). The volume of the bioreactor was 2.95 liters.
Sludge loading was kept stable at approximately 6 g/l by appropriate wasting of mixed liquor suspended solids (MLSS).
All chemicals used were reagent or HPLC grade materiels and distilled water was used for all solution preparation.

The vitamin and inorganic micronutriments solution was prepared as described in OECD guidelines. Except that all sulfate salts in the multiminerals solution were replaced with the corresponding chloride salts to help close the sulfur balance.

Remark : Frost (1991) identified that Escherichia coli K-12 bacteria can grow on MSA resulting in the complete mineralization of MSA to carbon dioxide and sulfate.

The literature further suggests that the ability to utilize sulfonates as a source of carbon and energy is applicable to many other microorganisms.

Methanesulfonate is used by diverse aerobic bacteria as a source of sulfur for growth, but is not known to be used by anaerobes either as sulfur source, a fermentation substrate, an electron acceptor.
MSA has been identified as a major photochemical oxidation product of DMS. DMS and MSA are predominantly biogenic in origin and are the main gaseous links in the biogeochemical sulfur cycle.

Source : ATOFINA, PARIS-LA-DEFENSE, FRANCE.

Test condition :

- Glass continuous culture (CC) Bioreactor:
The MSA bioreactor influent nutrient solution (per litre) was prepared by adding:
160 mg bacto beef extract, 110 mg bacto peptone, 90 mg bacto Urea, 0.6 ml ethanol and 0.6 ml methanol to the above OECD vitamin and inorganic micronutrient solution.
The MSA bioreactor feed solution's BOD/N/P ratio was 100/5.7/1.1.
- Pretreatment: no data

STOCK AND TEST SOLUTION AND THEIR PREPARATION
The MSA standard solution was prepared by weighing 1000 mg of MSA into 1 liter volumetric flask and diluting to the mark with distilled water.

MSA concentrations were increased by a factor of 2 from 5 to 2000 mg/L during the course of this study.In addition , the amount of sodium bicarbonate added to the MSA bioreactor solution for buffering was increased from 0.75 to 1.5 g/L with increasing MSA concentrations.

DILUTION WATER
- Source: Distilled water was used for all solution preparations.

pH: The pH was monitored throughout the study and was maintained in the range 6.8 to 7.8 through the addition of sodium bicarbonate.

TEST SYSTEM
- Exposure schedule: The hydraulic residence time (HRT) was maintained at approximately 60 hours. The optimum HRT for the microorganisms in the bioreactor was found to be >= 40 and <= 60 hours.

MONITORING OF TEST SUBSTANCE CONCENTRATION: IC analysis of MSA, chloride, phosphate, sulfate and nitrate was conducted using a Dionex DX300 ion chromatography system. The anions are separated using the HPIC-AS4A exchange separator column.

using the HPIC - AS4A exchange separator column.

Attached document : Magliette figure 2.bmp

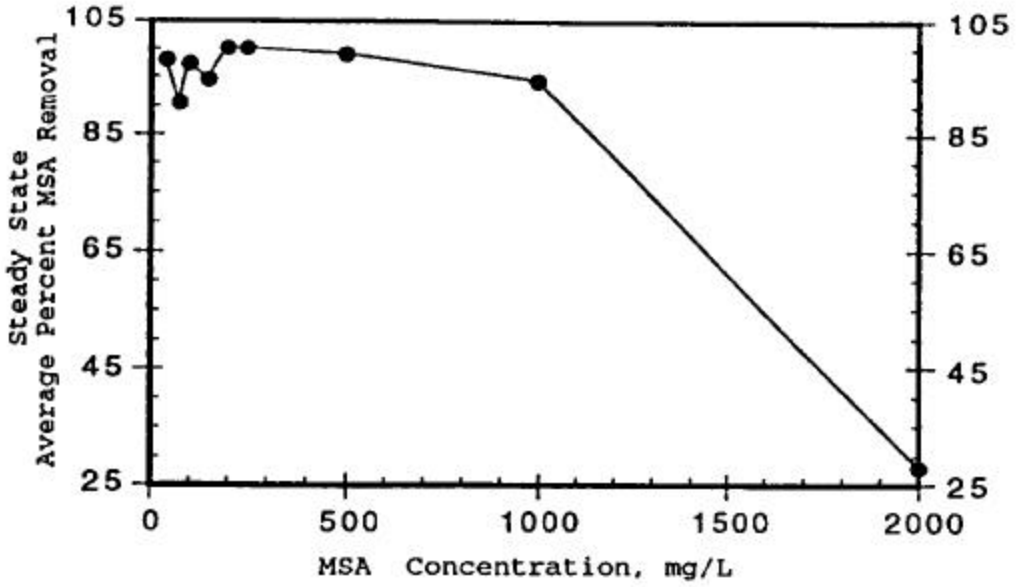


Figure 2. MSA removal as a function of MSA Concentration.

Conclusion : The activated sludge organisms were gradually challenged with increasing MSA concentrations (a factor of <=2). Rapid increase in MSA concentrations were found to be inhibitory. Higher MSA concentrations (2000 mg/L) were found to be inhibitory to the activated sludge organisms.

As long as the MSA concentration does not exceed 1000 mg/L, the sewering of MSA to a well acclimated activated sludge treatment plant should pose no acute adverse impact on it's performance.

These results point to the fact that MSA degraders are probably ubiquitous and abundant in nature.
Baker et al (1991) postulated that the sulfonic acid functional group of MSA must be available for microbial utilization. They also demonstrated that the methylotrophs and sulfur bacteria are able to derive energy from the degradation of MSA.

This study results indicate that microorganisms naturally present in any activated sludge wastewater treatment plant can be acclimated to degrade MSA.

Microorganisms play a vital role in the biogeochemical sulfur cycle.

Reliability : (4) not assignable
03.01.2003 (16)

Type : aerobic
Inoculum :

Remark : Methanesulfonate offers a number of hypothetical possibilities for use as a substrate for microbial growth and metabolism.
It could provide sulfur for biosynthesis to any organism able to cleave the C-S bond under aerobic or anaerobic conditions.
It could undergo oxidation to carbon dioxide and sulfate, yielding metabolic energy for growth from the oxidation of methyl-groups, and even of the sulfonate moiety to sulfate. Its methyl group could provide carbon intermediates for Methylotrophic bacteria.

intermediates for Methylotrophic bacteria.
It might be used anaerobically either as a terminal electron acceptor for anaerobic respiration, as a substrate for fermentative disproportionation or even as a substrate for methanogenesis from its methyl groups by archaeal methanogens.

Methanesulfonic acid is a very stable strong acid and a key intermediate in the biogeochemical cycling of sulfur.

Methane sulfonate oxidation is initiated by cleavage catalysed by methanesulfonate monooxygenase. Apart from very limited mention of mathanesulfonic acid as a sulphur source for some organisms, no work had at that time been done to investigate the microbial degradation of this important intermediate in the global sulphur cycle.

Methanesulfonate is remarkably stable. Such data indicate that the absence of detectable methanesulfonate from soils and the marine environment is because of its biological rather than chemical degradation. Methanesulfonate has thus had a continuous and significant imput to marine and terrestrial environments.Ample time was thus available for the evolution of bacteria able to use methanesulfonate, and the absence of measurable quantities from soils and water indicates its ready degradation by the modern microflora.

The methanesulfonate monooxygenase of Methylosulfonomonas and Marinosulfonomonas:

Cell-free extracts of strain M2 contained a cytoplasmic methanesulfonate monooxygenase activity that was specifically induced by growth on methanesulfonate and catalysed methanesulfonate-dependent NADH. The methanesulfonate monooxygenase of strain M2 was resolved into three distinct fractions, none with individual methanesulfonate oxidizing activity, but which together were reconstituted into an active form.

It has been possible to design gene-specific primers to analyse newly isolated methanesulfonate users
From a variety of different environments by using the sequences determined for the highly conserved msmA from Methylosulfonomonas strain M2 and Marinosulfonomonas strain TR3.
DNA was extracted directly from aerobic enrichment cultures, with methanesulfonic as the sole substrate, using inocula from soil, sediment and marine samples.

These observations strongly support the view that a conserved methanesulfonic monooxygenase enzyme is present in a variety of bacterial genera and consequently enables the use of molecular techniques for the direct study of environmental samples.

Source : ATOFINA, PARIS-LA-DEFENSE, FRANCE

Conclusion : -Methanesulfonate as a sulfur source
Numerous longer-chain sulfonates are used as sulfur sources for growth by diverse bacteria, fungi and algae. But the use of methanesulfonate seems to be more restricted.
Methanesulfonate is the most stable sulfonate and the poorest sulfur source. A number of aerobic bacteria can use methanesulfonate as a sulfur source.

-Methanesulfonate as a respiratory electron acceptor, fermentation substrate or methanogenic substrate.
To date no bacteria have been described that can degrade methanesulfonate in the absence of oxygen. But the possibility to find anaerobic methanesulfonate degraders cannot be excluded.
Methanesulfonate is an analogue of methanephosphonate, which had been

Methanesulfonate is an analogue of methanephosphonate, which had been shown to be degraded by Comamonas testosteroni to produce methane and phosphate by hydrolytic cleavage of the C-P bond.
Methanesulfonate has not yet been shown to be a respiratory electron acceptor for any sulfate-reducing bacterium. Use of methanesulfonate or its hydrolysis products as respiratory hydrogen acceptors by the action of methanogens or sulfate reducers could conceivably lead to its anaerobic mineralization to methane and sulfite or sulfide.

-Methylotrophic growth on methanesulfonate as a source of carbon and energy.
Desulfonation of methanesulfonic can lead initially only to methanol or formaldehyde. Thus only methylotrophic bacteria are likely to be able to use methanesulfonate as the sole substrate for growth as any non-methylotrophic heterotroph possessing a methanesulfonate monooxygenase enzyme would not be able to derive cell-carbon exclusively from methanesulfonate, although the oxidation of methanesulfonate to carbon dioxide and water could act as a supplementary energy source.
The first bacterium isolated on methanesulfonate as sole substrate was characterized as belonging to a novel genus within the a-proteobacteria, Methylosulfonomonas (soils).

These observations and the inability of some other well-known genera of methylotrophs to use methanesulfonate, suggested that methanesulfonate use might be restricted to these specialized genera.

-Mechanism of growth on methanesulfonate
All of the aerobic bacteria shown to date to use linear alkanesulfonate as growth substrates probably use a monooxygenase to split the C-S bond of the sulfonate. Hydrolysis of methanesulfonate to produce methanol does not occur in any of the methylotrophs isolated so far.

The observations strongly support the view that a conserved methanesulfonic monooxygenase enzyme is present in a variety of bacterial genera and consequently enables the use of molecular techniques for the direct study of environmental samples.

Reliability : (4) not assignable
30.12.2002 (17)

Remark : This paper describes the initial purification and characterization of an MSA-monooxygenase (MSAMO), capable of C-S bond fission in the presence of NADH, from methylotroph strain M2, and also the isolation of mutants of strain M2 lacking the ability to carry out such a reaction.

A major product of DMS oxidation reactions can be methanesulfonic acid MSA, a stable strong acid that does not undergo photochemical oxidation. Once deposited on the earth, MSA is thought to undergo biodegradation ultimately to form CO2 and SO42-, which in turn can be incorporated into DMSP(dimethylsulfoniopropionate), thus completing part of the biogeochemical organic sulfur cycle.
The stability of MSA is due mainly to the strength of the C-S bond found in all organosulfate compounds.

Aliphatic sulfonates have been also identified as sole sulfur sources for bacteria isolated from soil and sewage, as well as certain enteric bacteria and some yeasts.
Bacteria have also been isolated which can utilize both primary and secondary alkyl sulfonates as the only source of carbon and energy.

In both cases, primary biodegradation is proposed to be by direct desulfonation of the alkyl sulfonate. The mechanism proposed involves the

desulfonation of the alkyl sulfonate. The mechanism proposed involves the insertion of a hydroxyl group at the a-carbon by a monooxygenase enzyme.

Initial studies on the utilization of MSA as a sole source of carbon and energy by the methylotroph M2; Kelly have indicated that the initial step involves desulfonation to produce formaldehyde and sulfite.
Initial studies with cell-free extracts of strain M2 have identified an enzyme capable of oxidising MSA in the presence of NADH.
The enzyme is cytosolic and is induced in strain M2 only when grown on MSA as sole carbon and energy source. The enzyme had:
- pH optimum of 7-7.2
- and was stable for several days at -20°C
- no appreciable loss of activity was observed after 5 weeks at -70°C.

Result : MSAMO activity and inhibition in cell-free extracts
Initial studies with freshly prepared cell free extracts of MSA-grown strain M2 had revealed an enzyme capable of oxidising MSA in the presence of NADH.

Inhibition of the enzyme by metal chelators (bathophenanthroline, bathocuproine, neocuproine and sodium EDTA) indicated that the enzyme did indeed have associated metal ions, which are required for activity.
Electron transfer reactions were essential to the reactions producing cleavage of the C-S bond of MSA.

Isolations of mutants, which could not utilize methanol as sole carbon and energy source, but which were still able to utilize MSA, supports the proposed MSA oxidation pathway in strain M2, whereby MSA is oxidised directly to formaldehyde and sulfite, but methanol is not an intermediate in the pathway.
The ability of methanol mutants to grow at the expense of MSA confirmed that methanol was not an obligatory intermediate of the MSA degradative pathway in strain M2.

Elimination of methanol as a potential intermediate of MSA biodegradation and the production of sulfite by cell extracts of strain M2 is consistent with an oxygenolytic mechanism.

The ability of MSAMO to desulfonate only short-chain sulfonates suggests the metabolism of primary aliphatic sulfonates by different enzymes may be carbon-chain-length dependent.

The isolation of both structural and regulatory mutants of MSA metabolism facilitates their use in future-studies concerning the regulation of MSA oxidation pathways.
These double mutants could also be transport mutants lacking the ability to take up both MSA and formate into the cell.

Source : ATOFINA, PARIS -LA-DEFENSE, FRANCE.

Test condition : Growth of the organisms and preparation of cell-free extracts:
Methylotroph strain M2 was cultivated and maintained on mineral salts medium. Trace element solution and vitamin solution were added only when carbon substrates were added to give a final concentration of 20 mM. 15 g/l of Difco bacto agar was added to the medium prior to sterilization.

Initially a batch culture of strain M2 was grown in 5000 ml fermenter equipped with pH, oxygen and temperature control.
- Temperature: 30 °C
- pH: 7
- Air was supplied to the ferm enter continuously at 1 ml (ml culture -1) min -1

- Treatment: Cells were harvested by centrifugation (17000 g) at 4°C, washed three times with 40 mM Tris/HCl buffer. The cells were either drop-

washed three times with 40 mM Tris/HCl buffer. The cells were either drop-frozen in liquid nitrogen prior to storage at - 70°C or immediately broken by two or three passages through a chilled French pressure cell at 137 MPa. Cell debris was removed by centrifugation (50 000 g) for 75 min at 4°C to yield the cell-free extract which was immediately used, or drop-frozen in liquid nitrogen and stored at - 70°C.

- Analytical methods: The protein content of cell extracts was determined by the methods of Bradford. Analysis of cell extracts and protein fractions was carried out on 12% (W/V) acrylamide gel.

- Whole-cell oxygen electrode studies. Only 20-30 mg dry weights of washed cells were used in each experiment and the change in oxygen uptake was determined after the addition of 1 mM (final concentration) of each substrate.

- Enzyme assay: A modification of the spectrophotometer assay for measuring MSA monooxygenase activity was used, based on monitoring the substance-simulated oxidation of NADH at 340 nm where 2 nmol FAD and 100 nmol Fe(NH4)2(SO4)2 were also added to the reaction mixture. This assay was based on the detection of sulfite produced following MSA oxidation.

Mutagenesis: Was terminated after incubation with shaking (200 r.p.m) at 30°C for sufficient time (23-25 min). Washed cells were resuspended in 200 ml sterile Min E medium and allowed to recover by incubation with shaking at 30°C for 24 hours in the presence of vitamin solution, trace element solution, 20 mM each of MSA, methylamine, formate, and 0,2 % (v/v) methanol. The cells were then subjected to penicillin-enrichissement by centrifugation and resuspension in Min E medium.

Conclusion : The initial step in the biodegradative pathway of MSA in strain M2 involved an inducible NADH-specific monooxygenase enzyme (MSAMO). Analysis of mutants of strain M2 defective in the metabolism of C, compounds indicated that methanol was not an intermediate in the degradative pathway of MSA and also confirmed the involvement of a multicomponent enzyme in the degradation of MSA by methylotroph strain M2.

Reliability : (4) not assignable
30.12.2002

(18)

3.6 BOD5, COD OR BOD5/COD RATIO

3.7 BIOACCUMULATION

3.8 ADDITIONAL REMARKS

4.1 ACUTE/PROLONGED TOXICITY TO FISH

Type	:	static
Species	:	Oncorhynchus mykiss (Fish, fresh water)
Exposure period	:	96 hour(s)
Unit	:	mg/l
NOEC	:	= 56
LC50	:	= 73
Limit test	:	
Analytical monitoring	:	yes
Method	:	OECD Guide-line 203 "Fish, Acute Toxicity Test"
Year	:	1992
GLP	:	yes
Test substance	:	other TS
Remark	:	Study peer reviewed
Result	:	RESULTS: RANGE FINDING TEST: Prior to initiating the definitive study, a preliminary test was conducted at Springborn during which rainbow trout were exposed under static conditions to nominal concentrations of 0.10, 1.0, 10, 40 and 100 and a control. Mortality of 100% was observed among rainbow trout exposed to the 100 mg A.I./L treatment level following 24 hours of exposure. At test termination (96 hours), no mortality or adverse effects were observed among the rainbow trout exposed to the 0.10 to 40 mg A.I./L. treatments or the control. Based on these results, nominal concentrations of 13, 22, 37, 60 and 100 mg A.I./L were selected for the definitive study. In an effort to evaluate the effect of pH on the toxicity of the test substance to the exposed organisms, a duplicate set of exposure solutions was established at the 60 and 100 mg A.I./L treatment levels. The pH of these solutions was adjusted to 7.1 - 7.3 with sodium hydroxide. RESULTS: EXPOSED - Nominal/measured concentrations: Measured concentrations w ere within 86% of nominal - Mortality and effect data: 100% mortality at 96 mg/l group only - Concentration / response curve: See Attached Document 5
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	TEST ORGANISMS - Supplier: Springborn culture facility - Batch number: 97A71 - Wild caught: no - Post-hatch transfer time: no data - Age: no data - Size: 45 mm (range 40 to 54 mm) - Weight: 0.86 (range 0.57 to 1.43 g) - Feeding: food was stopped 24-h prior to test initiation - Pretreatment: Prior to testing, the fish were held in a 500-L fiberglass tank under a photoperiod of 16h light and 8h darkness for 14 days. No mortality was observed during the 48-h period prior to testing. - Feeding during test: No - Controls:Yes STOCK AND TEST SOLUTION AND THEIR PREPARATION - Dispersion: A 50 mg/ml stock solution was prepared by dissolving 35.36 g of methane sulfonic acid (from a 70% solution) in 500 ml of distilled water.

Treatment level solutions were prepared by adding the appropriate amount of the 50 mg A.I. (Active Ingredient)/ml to the 15L of dilution water.
- Vehicle, solvent: water (the same as that used in the fish holding tank)
- Concentration of vehicle/solvent: useless data
- Other procedures: none

STABILITY OF THE TEST CHEMICAL SOLUTIONS:
Nothing to report.

REFERENCE SUBSTANCE:
None

DILUTION WATER
- Source: from a 100-meter deep bedrock well and well water supplied by the Town of Wareham, Massachusetts.
- Aeration: yes
- Alkalinity: 40 mg/l (CaCO3)
- Hardness: 34 mg/l (CaCO3)
- Salinity: 10 mg/l
- TOC (Total Organic Carbon): 0.57 mg/l
- TSS: 0.0 mg/l
- pH: 7.3
- Oxygen content: 76 to 86%
- Conductance: 100 to 145 µmhos/cm

TEST SYSTEM
- Concentrations: 13, 22, 37, 60 and 100 mg A.I./L
- Renewal of test solution: no
- Exposure vessel type: 18.9 -L glass aquarium, containing 15L of test solution
- Number of replicates: 2
- Number of individuals per replicate: 10
- Test temperature: 12°C
- pH: cf. Attached document 1
- Adjustment of pH: In order to evaluate the effect of pH on the toxicity of the substance, a duplicate set of exposure solutions was established at the 60 and 100 mg A.I./L treatment level. The pH of these solutions was adjusted to 7.1 to 7.3 with a 4.0 NaOH solution.
- Intensity of irradiation: 80 footcandles at the solution's surface
- Photoperiod: 16h light / 8h darkness

ENDPOINTS ASSESSED:
Fishes: mortality (dead fishes were removed twice a day), sublethal effects, LC50 and NOEC
Water: physical observations (precipitate, film on surface,¿), pH, dissolved oxygen, temperature, total alkalinity, methane sulfonic acid concentration (titrated at 0 and 96 hours in all replicate solutions)

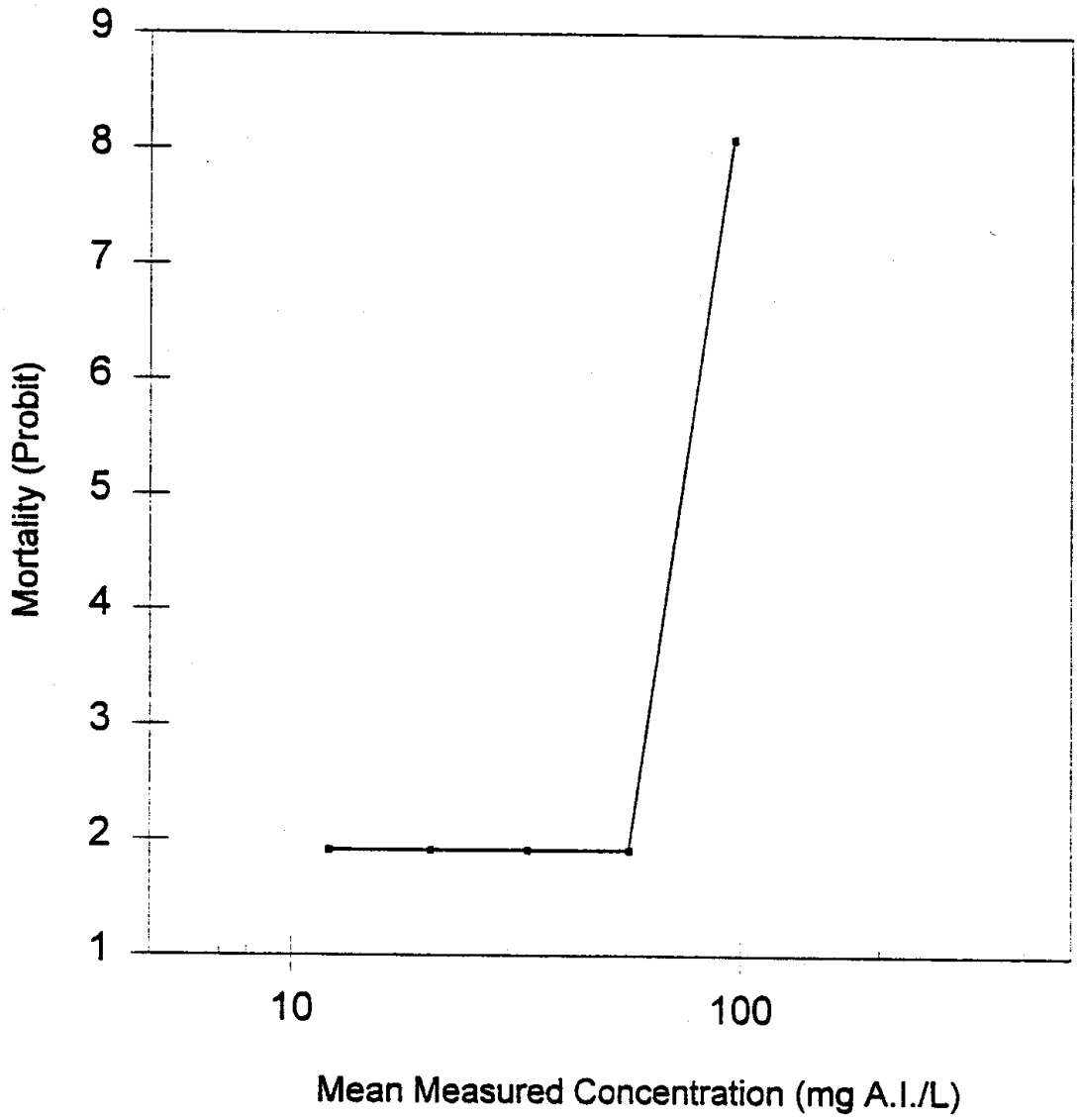
MONITORING OF TEST SUBSTANCE CONCENTRATION:
see "Endpoints assessed".
Analytical device: HPLC

STATISTICAL TEST:
Moving average angles, Probit analysis

Test substance : Source: Elf Atochem
Batch number: 2324E20HD1
Purity: 70.3%

Attached document : 75-75-2 Acute tox. to fish (97-4-7192) Dose-response curve.bmp

The 96-hour concentration response (mortality) curve for rainbow trout (*Oncorhynchus mykiss*) to exposed to methane sulfonic acid under static conditions.



Conclusion : The 96-hour LC50 was esimated by nonlinear interpolation to be 73 mg A.I./L with a 95% confidence interval, calculated by binomial probability to be 56 to 96 mg A.I./L.
The NOEC was determined to be 56 mg A.I./L.
No mortality occured in the 62 and 89 mg A.I./L solutions which were adjusted to pH 7.1 to 7.3.

Reliability : (1) valid without restriction

Flag	:	Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

03.01.2003 (19)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

Type	:	static
Species	:	Daphnia magna (Crustacea)
Exposure period	:	48 hour(s)
Unit	:	mg/l
NOEC	:	= 120
EC50	:	= 260
Analytical monitoring	:	yes
Method	:	OECD Guide-line 202
Year	:	1981
GLP	:	yes
Test substance	:	other TS
Method	:	- Analytical device: General Electric type 217 light-meter, Spring Instrument Company (YSI) model 33 salinity-conductivity-temperature-meter, Hanna model HI9024 pH-meter, YSI dissolved oxygen-meter, Fisher Scientific Min-Max thermometer, HPLC. - Statistical test: Moving average angle and probit analysis
Remark	:	Study peer reviewed
Result	:	Based on measured concentrations, the 48-hour EC50 value was estimated by nonlinear interpolation to be 260 mg A.I./L with 95% confidence limits calculated by binomial probability of 210 to 330 mg A.I./L. The No-Observed-Effect Concentration (NOEC) through 48 hours was determined to be 120 mg A.I./L. Immobilization in the 620 and 900 mg A.I./L solutions which were adjusted to pH 7.9 to 8.3 were similar to that of the control. These data indicate that the acidic nature of the non pH adjusted test solution probably contributed to the immobilization observed during the definitive test.
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	TEST ORGANISMS - Source/Supplier: Springborn Laboratories culture facility - Breeding method: no specific data - Age: < 24 hours - Feeding: Ankistrodesmus falcatus (unicellular green algae) 2ml of a 4.10E7 cell/ml suspension per day in each vessel. - Pretreatment: no data - Feeding during test: no - Control group: yes STOCK AND TEST SOLUTION AND THEIR PREPARATION - Dispersion: a 50 mg A.I./L stock solution was prepared by diluting 7.1124 g of the 70% solution of methane sulfonic acid with distilled water up to 100 ml. - Vehicle, solvent: water - Concentration of vehicle/solvent: no data STABILITY OF THE TEST CHEMICAL SOLUTIONS: no data REFERENCE SUBSTANCE: no DILUTION WATER - Source: fortified well water and filtered through an Amberlite XAD-7 resin column

- Composition:
Total hardness as CaCO3: 170 mg A.I./L
Total alkalinity as CACO3: 120 mg A.I./L

PRELIMINARY STUDY: yes, range-finding study

TEST SYSTEM
- Concentrations: 120, 210, 330, 570 and 890 mg A.I./L
- Renewal of test solution: no
- Exposure vessel type: 1-litre glass beaker, containing 500 ml of test solution
- Number of replicates: 2
- Number of individuals per replicate: 5
- Test temperature: 19 to 21°C
- Dissolved oxygen: 8.0 to 8.2 mg/l
- pH: 8.3
- Adjustment of pH: between 7.9 and 8.3, only for a duplicate set including 600 and 1000 mg A.I./L
- Intensity of irradiation: 60 footcandles at the solution surface
- Photoperiod: 16 hours of light + 8 hours darkness

DURATION OF THE TEST: 24 and 48 hours

QUALITY CRITERIA:
- Control mortality (OK if < 10%):
- pH (OK if equals to initial pH ± 1): not fulfilled in the 330 mg/l flask
- Dissolved oxygen (OK if > 2mg/L): OK
- Test substance concentration (OK if > 80% of initial concentration): OK

MONITORING OF TEST SUBSTANCE CONCENTRATION:
One water sample was removed from both replicate solutions of each treatment level and the controls at 0 and 48 hours for the analysis of methane sulfonic acid concentration.

Test substance : Source: Elf Atochem
Batch number: 2324E20HD1
Purity: 70.3%

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

30.12.2002 (20)

Type :
Species : Daphnia magna (Crustacea)
Exposure period : 24 hour(s)
Unit : mg/l
EC50 : = 1.7
EC100 : = 7.405 calculated
Analytical monitoring : no
Method : ISO 6341 15 "Water quality - Determination of the inhibition of the mobility of Daphnia magna Straus (Cladocera, Crustacea)"
Year :
GLP : no
Test substance : as prescribed by 1.1 - 1.4

Remark : Study peer reviewed
Result : NOEC: undetermined
EC50: 1.7 mg/l
EC100: 7.405 mg/l

EC50 with potassium dichromate: 1.0 mg/l

Source : ELF ATOCHEM Paris la defense 10
EUROPEAN COMMISSION - European Chemicals Bureau Ispra (VA)

Test condition :

TEST ORGANISMS
- Source/Supplier: no data
- Breeding method: synthetic medium
- Age: no data
- Feeding: (Scenedesmus subspicatus) + Tetramin
- Pretreatment: no data
- Feeding during test: algae
- Control group: yes

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- Dispersion: no data
- Vehicle, solvent: water
- Concentration of vehicle/solvent: no data
- Other procedures: no data

STABILITY OF THE TEST CHEMICAL SOLUTIONS: no data

REFERENCE SUBSTANCE: potassium dichromate

DILUTION WATER
- Source: no data
- Composition: no data

PRELIMINARY STUDY: no data

TEST SYSTEM
- Concentrations: no data
- Renewal of test solution: no data
- Exposure vessel type: no data
- Number of replicates: no data
- Number of individuals per replicate:
- Test temperature: no data
- Dissolved oxygen: 9.0 mg/l
- pH: 3.51
- Adjustment of pH: no data
- Intensity of irradiation: no data
- Photoperiod: no data

QUALITY CRITERIA:
- Control mortality (OK if < 10%): no data
- pH (OK if equals to initial pH ± 1): no data
- Dissolved oxygen (OK if > 2mg/L): OK
- Test substance concentration (OK if > 80% of initial concentration): no data

SAMPLING: no data

MONITORING OF TEST SUBSTANCE CONCENTRATION: no data

Test substance : Source: Atochem
Batch number: 1228/89
Purity: no data

Reliability : (2) valid with restrictions Scarce data do not allow to give a better reliability index.
Scarce data do not allow to give a better reliability index.

03.01.2003 (21)

Type : static
Species : Daphnia pulex (Crustacea)
Exposure period :
Unit :

Remark : Study peer reviewed

Result	:	Average tolerance level, 24h = 33 mg/l Average tolerance level, 48h = 12 mg/l
Source	:	Atofina, Paris-la-Défense, France
Test condition	:	Werner's method was applied (WERNER, A.E., 1963. Sulphur compounds in kraft pulp mill effluents.Can. Pulp Paper Ind., 16, 3, 35-43). The tests were made in glass cylindres of 110 ml capacity. Volume of test solution : 100 ml. Temperature : About 20°C (The test was done at Room temperature). Conditions : The water fleas were so handled that no air could penetrate beneath their back shell during transfer. Concentrations: The dilutions required were made in such a way that the test-animals would not suddenly encounter higher concentrations than were desired.
Reliability	:	(4) not assignable Documentation insufficient for assessment. Documentation insufficient for assessment.

03.01.2003 (22)

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

Species	:	Selenastrum capricornutum (Algae)
Endpoint	:	growth rate
Exposure period	:	96 hour(s)
Unit	:	mg/l
NOEC	:	= 5.8 measured/nominal
EC10	:	= 2.1 - 15 calculated
EC50	:	= 7.2 - 20 calculated
EC90	:	= 12 - 26 calculated
EbC50	:	= 8.6 - 19 calculated
ErC50	:	= 7.6 - 24 calculated
Limit test	:	no
Analytical monitoring	:	yes
Method	:	OECD Guide-line 201 "Algae, Growth Inhibition Test"
Year	:	1984
GLP	:	yes
Test substance	:	other TS
Remark	:	Study Peer reviewed.
Result	:	Measured concentrations were at least 87% of nominal concentrations. Percent cell density; biomass; growth inhibition 2.7 mg/l (0.62%; -5.6%; -1.7%) 5.8 mg/l (5.4%; 7.0%; 0.38%) 12 mg/l (7.9%; 14%; 0.99%) >24 mg/l (100%; 100%; 100%) Percent cell density; biomass; growth inhibition when pH adjusted to 7.5 49 mg/l (0.62%; -5.6%: -1.6%) 95 mg/l (2.1%; 1.6%; -0.76%) Based on cell density, the NOEC was determined to be 5.8 mg A.I./L. The 96-hour EC50, EC10 and EC90 were calculated to be 14, 8.8 and 19 mg A.I./L, respectively. Based on total biomass, the NOEC was determined to be 5.8 mg A.I./L. The 0-72 hour EbC50 was calculated to be 14 (8.6-19) mg A.I./L.

Based on average growth rate, the NOEC was determined to be 12 mg A.I./L. The 0-72-hour ErC50 was calculated to be 16 (7.6-24) mg A.I./L.

Source : Atofina, Paris-la-Défense, France

Test condition : TEST ORGANISMS
- Strain: 1648, class Chlorophyceae.
- Source/supplier: The alga was originally obtained from the University of Texas and maintained in stock sulture at springborn.
- Method of cultivation: The stock cultures were maintained within the following conditions: a shaking rate of 100+/-10 rpm, a temperature of 24+/-1°C and continuous illumination at the surface of the medium.
- Pretreatment: the inoculum used to initiate the toxicity test with methane sulfonic acid was taken from a stock culture that had been transferred to fresh medium three days before testing.
- Initial cell concentration: 10000 cells/ml.

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- Dispersion: The 100 mg A.I(active ingredient)/L test solution was prepared by adding 0.4267 g of methane sulfonic acid to 3000-ml of AAP medium.The resulting test solution was clear, no visible signs of undissolved test substance.
STABILITY OF THE TEST CHEMICAL SOLUTIONS: No data.

DILUTION WATER
Algal assay procedure (AAP) medium.
GROWTH/TEST MEDIUM CHEMISTRY
$NaNO_3$: 25.5 mg/l
$MgCl_2.6H_2O$: 12.16 mg/l
$CaCl_2.2H_2O$: 4.41 mg/l
$MgSO_4.7H_2O$: 14.7 mg/l
$K_2HPO_4.3H_2O$: 1.368 mg/l
$NaHCO_3$: 15 mg/l
H_3BO_3 : 185.5 µg/l
Na_2SeO_4 : 1.88 µg/l
$MnCl_2.4H_2O$: 415.4 µg/l
$ZnCl_2$: 3.270 µg/L
$CoCl_2.6H_2O$: 1.43 µg/l
$CuCl_2.2H_2O$: 0.012 Mg/l
$Na_2MoO_4.2H_2O$: 7.26 µg/L
$FeCl_3.6H_2O$: 159.8 Mg/L
$Na_2EDTA.2H_2O$: 300 Mg/L
- pH: The pH of the culture medium was adjusted, if necessary, to pH 7.5+/-0.1 with either 0.10 N HCl or 0.10N NaOH.

TEST SYSTEM
- Test type: The test was conducted in an environmental chamber (adjusted to maintain the test conditions) with an orbital shaker table.
- Concentrations:
- Renewal of test solution: no.
- Exposure vessel type: Replicate sterile 250-ml Erlenmeyer flasks were conditioned prior to use by rinsing with the appropriate exposure solution. All test vessels were fittes with stainless steel caps to permit gas exchange.
- Number of replicates: 3.
- Concentrations:
Nominal test concentration: 3.1, 6.3, 13, 25, 50, and 100 mg A.I./L; 50 and 100 mgA.I./L adjusted to pH 7.5.
Mean measured concentration: 2.7, 5.8, 12, 24, 47 and 96 mg A.I./L; 49 and 95 mg A.I./L respectively.
- Test temperature: 24+/-1°C.
- pH: From 7.6 to 7.5 at T0 and 9.4 to 9.3 at 96 hours.
- Photoperiod: Continuous light intensity of 3200 to 5400 lux.

TEST PARAMETER: Inhibition of test density (inhibition of cell

TEST PARAMETER: Inhibition of test density (inhibition of cell multiplication)

MONITORING OF TEST SUBSTANCE CONCENTRATION: Due to the acidic nature of the test solutions, two additional concentrations, 50 and 100 mg A.I./L were adjusted to pH 7.5 and tested in addition to the above concentrations.

Test substance : -Source: ATOCHEM
-Batch number: 2324E20HD1
-Purity: 70.3%
-Water solubility: Miscible in water.

Attached document : 75-75-2 5.bmp

Table 6. Calculated biomass (area under the growth curve) of *Selenastrum capricornutum* after 24, 48 and 72 hours of exposure to methane sulfonic acid.

Mean Measured Concentration (mg A.I./L)		Biomass (x 10^4 cells·days/mL) Observation Interval 0 - 24 Hour	24 - 48 Hour	48 - 72 Hour	Total Area 72 Hour	72 Hour Percent Inhibition[a]
Control	A	2.7	14	34	51	
	B	3.5	14	32	50	
	C	2.1	12	34	48	
	Mean(SD)[b]	2.8(0.69)	14(1.4)	33(1.3)	50(1.6)	NA[c]
2.7	A	3.2	14	35	53	
	B	4.1	15	36	56	
	C	2.1	13	34	49	
	Mean(SD)[b]	3.2(0.99)	14(1.1)	35(1.0)	53(3.1)	-5.6
5.8	A	2.7	13	34	50	
	B	2.6	11	30	43	
	C	1.9	12	32	46	
	Mean(SD)[b]	2.4(0.47)	12(0.99)	32(2.2)	46(3.2)	7.0
12	A	1.6	10	32	43	
	B	2.5	12	30	44	
	C	1.5	9.8	29	40	
	Mean(SD)[b]	1.9(0.54)	10(0.95)	30(1.2)	43(2.1)[d]	14
24	A	-0.50	-1.0	-0.96	-2.5	
	B	-0.37	-0.75	-0.60	-1.7	
	C	-0.37	0.88	-0.84	-2.1	
	Mean(SD)[b]	-0.42(0.072)	-0.88(0.13)	-0.80(0.18)	-2.1(0.37)[e]	100
47	A	-0.50	-0.88	-0.84	-2.2	
	B	-0.50	-1.0	-0.84	-2.3	
	C	-0.50	-0.88	-0.72	-2.1	
	Mean(SD)[b]	-0.50(0.00)	-0.92(0.072)	-0.80(0.069)	-2.2(0.12)[e]	100
96	A	-0.50	-0.75	-0.72	-2.0	
	B	-0.37	-0.75	-0.72	-1.8	
	C	-0.50	-0.1.0	-0.96	-2.5	
	Mean(SD)[b]	-0.46(0.072)	-0.83(0.14)	-0.80(0.14)	-2.1(0.33)[e]	100

[a] Percent inhibition relative to the control.
[b] Mean, standard deviation (SD) and percent inhibition were calculated from original raw data, not from the rounded values presented in this table.
[c] NA = not applicable
[d] Significantly reduced as compared to the control based on Williams' Test.
[e] Concentration excluded from statistical analysis because negative values can not be analyzed.
[f] The pH of the test solution was adjusted to pH 7.5 prior to the addition of the test organism.

Conclusion : Based on the results of definitive test, it is evident that the acidic nature of the test substance contributed to the toxicity observed. Additionally, the inhibition of algal growth in the acidic medium at a nominal concentration of 25 mg A.I./L was reversible once diluted to a nontoxic pH.

Cell densities in the 49 and 95 mg A.I./L solutions which were adjusted to

Cell densities in the 49 and 95 mg A.I./L solutions which were adjusted to pH 7.5 were similar to that of the control. This data indicates that the acidic nature of the non-pH adjusted test solutions was probably the cause of algal inhibition in the definitive test.

The results of the recovery phase indicated that the effects of Methane sulfonic acid on algal growth were algistatic rather than algicidal.

Reliability : (1) valid without restriction
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

03.01.2003 (23)

4.4 TOXICITY TO MICROORGANISMS E.G. BACTERIA

Type : other: activated sludge
Species : activated sludge of a predominantly domestic sewage
Exposure period : 3 hour(s)
Unit : mg/l
EC50 : = 530
Analytical monitoring : no
Method : ISO 8192 "Test for inhibition of oxygen consumption by activated sludge"
Year : 1986
GLP : no data
Test substance :

Method : Analytical device: Jenway 3410 (oxymeter, conductimeter, pH-meter)
Remark : Study peer reviewed
Result : See Attached Document

At the highest concentration (1000 m/l) methane sulfonic acid exhibited a 76.2% inhibiting effect on activated sludge respiration after a 3-hour exposition.

The lowest tested concentration (100 mg/l) did not reveal any inhibiting effect (1.2%).

Source : Atofina, Paris-la-Défense, France
Test condition : TEST ORGANISMS
- Source/supplier: Versailles water waste treatment plant
- Laboratory culture: activated sludge is sampled then its suspended particles are titrated (result: 1.8 g/l). After centrifugation, supernatant is discarded and residue is resuspended in an isotonic medium mixed with synthetic medium (3:100 v/v) in order to have a 1.5 g/l final concentration in test flasks.
- Method of cultivation: no data
- Plate composition: no data
- Pretreatment: no data

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- Dispersion: a stock solution of 5 g/l methane sulfonic acid is prepared, then properly diluted
- Vehicle, solvent: ultrafiltered water

STABILITY OF THE TEST CHEMICAL SOLUTIONS: No data

REFERENCE SUBSTANCE: 3,5-dichlorophenol

DILUTION WATER
- Source: no data
- Aeration: yes

GROWTH TEST MEDIUM CHEMISTRY
. Synthetic medium:
Peptone: 16 g
Meat extract: 11 g
Urea: 3 g
NaCl: 0.7 g
CaCl2, 2H2O: 0.4 g
MgSO4, 7H2O: 0.2 g
K2HPO4: 2.8 g
Ultra-pure water: qsp 1000 ml

. Isotonic solution:
NaCl: 5 g
MgSO4, 7H2O: 0.12g
Ultra-pure water: qsp 1000 ml

- pH: 7.5 ± 0.5

TEST SYSTEM
- Exposure vessel type: 1 litre Erlenmeyer flasks
- Microorganism initial concentration: 1.5 g/l
- Exposure schedule: after 3 hours incubation, oxygen is measured every minute for approximately 10 minutes, until oxygen concentration reaches 1 mg/l
- Number of replicates per dose: 1
- Concentrations: 100, 180, 320, 580 and 1000 mg/l
- Control: vehicle
- Test temperature: 18.6 to 19.1°C
- Photoperiod: no data

TEST PARAMETER: Oxygen is measured in each test and control flask.

MONITORING OF TEST SUBSTANCE CONCENTRATION: no data

Test substance : Source: Elf Atochem
Batch number: 2690
Purity: 70%

Attached document : 75-75-2 Sludge respiration inhibition (dose table).bmp
75-75-2 Sludge respiration inhibition (results table).bmp

		Temps d'aération			
		30 minutes *		3 heures	
	Conc. (mg/l)	Taux de consommation ($mgO_2/l/h$)	Inhibition (%)	Taux de consommation ($mgO_2/l/h$)	Inhibition (%)
Blancs					
FB1	-	nd	-	52,8	-
FB2	-	nd	-	48,0	-
Moyenne	-	nd	-	50,4	-
Substance de référence : 3,5 dichlorophénol					
FR1	5	nd	nd	48,0	4,8
FR2	16	nd	nd	19,8	60,7
FR3	50	nd	nd	18,0	64,3
ACIDE METHANE SULFONIQUE					
FT1	100	nd	nd	49,8	1,2
FT2	180	nd	nd	30,0	40,5
FT3	320	nd	nd	37,8	25,0
FT4	580	nd	nd	28,2	44,0
FT5	1000	nd	nd	12,0	76,2

* nd : non déterminé

Conclusion : Methane sulfonic acid exhibit an inhibiting effect on oxygen consumption of microorganisms contained in activated sludge of a waste water treatment plant.
In the experimental conditions of this study, methane sulfonic acid presents a EC50-3h of 530 mg/l.

No physico-chemical oxygen consumption has been noticed.

Reliability : (1) valid without restriction
Flag : Material Safety Dataset, Directive 67/548/EEC
30.12.2002 (24)

Type : other
Species : Pseudomonas putida (Bacteria)
Exposure period : 16 hour(s)
Unit : mg/l
EC0 : calculated
EC10 : = .54 calculated
EC50 : = 1.8
Analytical monitoring : no data
Method : other: ISO 10712
Year :
GLP : no data
Test substance : other TS

Method : Analytical device: Perkin-Elmer lambda 5 spectrometer
Data treatment: Probit's method
Remark : Study peer reviewed
Result : Test substance results:
See attached document.

See attached document.

Reference substance results:
EC10 and EC50 of 3,5-dichlorophenol were respectively 8.1 and 13 mg/l.

Validity criterion:
Control growth factor = 218 (>60)

Test condition : TEST ORGANISMS
- Source/supplier: Institut Pasteur 103281
- Laboratory culture: no data
- Method of cultivation: no data
- Plate composition: no data
- Pretreatment: no data

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- Dispersion: no data
- Vehicle, solvent: test medium

STABILITY OF THE TEST CHEMICAL SOLUTIONS: No data

REFERENCE SUBSTANCE: 3,5-dichlorophenol

DILUTION WATER
- Source: no data
- Aeration: no data

GROWTH TEST MEDIUM CHEMISTRY
- no data
- pH: 3.25 to 4.01

TEST SYSTEM
- Exposure vessel type: no data
- Microorganism initial concentration: no data
- Exposure schedule: no data
- Number of replicates per dose: 3
- Concentrations: 0.25, 0.50, 1, 2, 4 and 8 mg/l
- Control: vehicle
- Test temperature: 21 ± 1°C
- Photoperiod: no data

TEST PARAMETER: turbidity is measured by a spectrophotometer at 436 nm

MONITORING OF TEST SUBSTANCE CONCENTRATION: no data

Test substance : Source: Elf Atochem
Batch number: no data
Purity: no data

Conclusion : Growth inhibition test was performed on Pseudomonas putida for 16 hours. Inhibition was assessed by a turbidimetric comparison between test and control cultures. Results were found as follows:
EC50 = 1.8 mg/l (IC95%: 1.2-2.6)
EC10 = 0.54 mg/l (IC95%: 0.23-0.85)

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC

03.01.2003 (25)

4.5.1 CHRONIC TOXICITY TO FISH

4.5.2 CHRONIC TOXICITY TO AQUATIC INVERTEBRATES

4.6.1 TOXICITY TO SEDIMENT DWELLING ORGANISMS

4.6.2 TOXICITY TO TERRESTRIAL PLANTS

4.6.3 TOXICITY TO SOIL DWELLING ORGANISMS

4.6.4 TOX. TO OTHER NON MAMM. TERR. SPECIES

Species	:	other avian: Agelaiues phoeniceus
Endpoint	:	mortality
Exposure period	:	
Unit	:	mg/kg bw
LC50	:	> 100
Method	:	other
Year	:	
GLP	:	
Test substance	:	
Source	:	Atofina, Paris-la-Défense, France
Reliability	:	(2) valid with restrictions Collection of data

30.12.2002 (26)

4.7 BIOLOGICAL EFFECTS MONITORING

4.8 BIOTRANSFORMATION AND KINETICS

27.12.2002

4.9 ADDITIONAL REMARKS

5.0 TOXICOKINETICS, METABOLISM AND DISTRIBUTION

5.1.1 ACUTE ORAL TOXICITY

Type	:	LD50
Value	:	380.8 - 1105.1 mg/kg bw
Species	:	rat
Strain	:	Sprague-Dawley
Sex	:	male/female
Number of animals	:	30
Vehicle	:	other: undiluted
Doses	:	300, 500 and 750 mg/kg
Method	:	other: EPA 40 CFR Part 792
Year	:	
GLP	:	yes
Test substance	:	other TS: anhydrous methanesulfonic acid

Remark : Study peer reviewed

Result : MORTALITY:
cf. Attached Document

CLINICAL SIGNS:
The majority of clinical abnormalities were observed in both the animals that died and those that survived to day 14 and included salivation, decreased activity, wobbly gait, breathing abnormalities, apparent hypothermia, decreased/no defecation, feces small in size, urine stain, hunched posture, unkempt appearance, rough haircoat, piloerection, extremities pale in color, dehydration, emaciation, distended abdomen, decreased food consumption and dark material around the facial area.

BODY WEIGHT:
Body weight loss was noted for one 300 mg/kg male, one 500 mg/kg male, two 500 mg/kg females and one 750 mg/kg male during the study day 0-7 body weight interval. Body weight loss was noted for one 300 mg/kg male and one 750 mg/kg male during the study day 7-14 body weight interval. Body weight gain was noted for all other surviving animals during the test period.

NECROPSY FINDINGS:
Gross internal findings observed in the animals that died included distension/abnormal content/reddened mucosa in the digestive tract, dark red/mottled lungs, blackish-purple spleens, dark red lymph nodes, stained glandular mucosa in the stomach and body fat depletion/discoloration/adhesions in the abdominal cavity. Gross internal findings observed at necropsy on study day 14 included abnormal content in the digestive tract and thickened mucosa in the stomach.

POTENTIAL TARGET ORGANS:
no specific target-organs were detected

SEX-SPECIFIC DIFFERENCES:
Whereas male LD50 is greater than 750 mg/kg, female LD50 has been valued at 461.2 mg/kg (IC95%: 313.3 - 679.0).

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Charles River Laboratories, Inc., Portage, Michigan.
- Age: no data
- Weight at study initiation: 200 to 300 g

- Weight at study initiation: 200 to 300 g
- Number of animals: 5 males + 5 females / dose
- Controls: no
- Other:

ADMINISTRATION:
- Exposure route: gavage
- Volume administered: < 20 ml/kg
- Post dose observation period: 14 days

EXAMINATIONS: clinical observations, body weight and necropsy

Test substance : Source: Elf Atochem
Batch number: 2690J24HS1
Purity: no data

Attached document : 75-75-2 Acute oral tox 3255.144.bmp

SLI STUDY NO.: 3255.144
CLIENT: ELF ATOCHEM

TABLE 1
AN ACUTE ORAL TOXICITY STUDY IN RATS
SUMMARY OF MORTALITY

PAGE 1

	Dose Level	No. of	Study Day															
Sex	(mg/kg)	Animals	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Mortality
Male	300	5	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	1/5
	500	5	0	0	0	0	0	0	0	0	1	1	0	0	0	0	0	2/5
	750	5	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1/5
Female	300	5	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1/5
	500	5	0	0	1	1	0	0	0	0	0	1	0	0	0	0	0	3/5
	750	5	1	0	0	0	0	1	1	1	0	0	0	0	0	0	0	4/5

Conclusion : Under the conditions of this test, the acute oral LD50 of Methane Sulfonic Acid, Anhydrous in the male rat was estimated to be greater than 750 mg/kg. In the female rat, the acute oral LD50 was determined to be 461 mg/kg. In the sexes combined, the acute oral LD50 was determined to be 649 mg/kg (IC95%: 380.8 -1105.1 mg/kg).

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

18.12.2002 (27)

Type : LD50
Value : 748 - 1791 mg/kg bw
Species : rat
Strain : Sprague-Dawley
Sex : male/female
Number of animals : 30
Vehicle : water
Doses : 500, 1000,1500 mg/kg
Method : other: EPA 40 CFR Part 792
Year :
GLP : yes
Test substance : other TS: 70% methanesulfonic acid

Method : Statistical test: LD50 was determined separa tely for both sexes, using Litchfield and Wilcoxon method.
Remark : Study peer reviewed
Result : MORTALITY:
cf. Attached Document

CLINICAL SIGNS:
The most notable clinical abnormalities observed during the study included salivation, breathing abnormali ties, wobbly gait, decreased activity, decreased defecation, rough haircoat, urine/fecal stain and dark material around the facial area.

BODY WEIGHT:
Body weight loss was noted for one 1000 mg/kg male and one 1000 mg/kg female during the study day 0-7 body weight interval and for one 300 mg/kg male and one 1000 mg/kg male during the study day 7-14 body weight interval. Body weight gain was noted for all other surviving animals during the test period.

NECROPSY FINDINGS:
The most notable gross internal findings were observed in the animals that died and included abnormal content and reddened/thickened mucosa and discoloration in the digestive tract, dark red foci on the liver and blackish-purple spleens. No significant gross findings were observed at necropsy on day 14.

POTENTIAL TARGET ORGANS:
Digestive tract

SEX-SPECIFIC DIFFERENCES:
Whereas male LD50 is 860.1 mg/kg (IC95%: 540.1 - 1369.7), female LD50 has been valued at 2407.6 mg/kg (IC95%: 944.2 - 6139.2).

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Charles River Laboratories, Inc., Portage, Michigan.
- Age: no data
- Weight at study initiation: 205,3 g (males) and 204.7 g (females)
- Number of animals: 5 males + 5 females / dose
- Controls: no

ADMINISTR ATION:
- Exposure route: gavage
- Volume administered: 0.38, 0.75 and 1.13 ml/kg
- Post dose observation period: 14 days

EXAMINATIONS: clinical observations, body weight, mortality and necropsy

Test substance : Source: Elf Atochem
Batch number: D11HD1
Purity: no data

Attached document : 75-75-2 Acute oral tox 3255.111.bmp

SLI STUDY NO.: 3255.111
CLIENT: ELF ATOCHEM

TABLE 1
AN ACUTE ORAL TOXICITY STUDY IN RATS
SUMMARY OF MORTALITY

Sex	Dose Level (mg/kg)	No. of Animals	Study Day 0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Mortality
Male	500	5	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1/5
	1000	5	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3/5
	1500	5	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	4/5
Female	500	5	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1/5
	1000	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0/5
	1500	5	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	3/5

Conclusion : Under the conditions of this test, the acute oral LD50 of 70% METHANE SULFONIC ACID in the male rat was determined to be 860.1 mg/kg (IC95%: 540.1-1369.7).
In the female rat, the oral LD50 was determined to be 2407.6 mg/kg (IC95%: 944.2-6139.2). Nothing explains such a sex-difference.
In the sex combined, the oral LD50 was valued at 1157.5 mg/kg (IC50%: 748.0-1791.0).

CAUTION: previous data are expressed in mg of tested solution (diluted at 70%). Results in terms of active substance can be obtained by mulptiplication by 0.7, as follows:
Males LD50: 602.07 mg/kg (IC95%: 378.07-958.79)
Females LD50: 1685.32 mg/kg (IC95%: 660.94-4297.44)
Global LD50: 810.25 mg/kg (IC95%: 523.6-1253.77)

Reliability : (1) valid without restriction
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

18.12.2002 (28)

Type : LD50
Value : = .146 - .54 ml/kg bw
Species : rat
Strain : Wistar
Sex : male
Number of animals : 28
Vehicle : other: undiluted
Doses : 0.25, 0.5 and 1.0 ml/kg (methanesulfonic acid)
2,4,8 g/kg (methanesulfonic acid sodium salt)
Method : other: unprecised EPA guidance
Year : 1975
GLP : no
Test substance : other TS

Remark : Study peer reviewed
Result : CLINICAL OBSERVATIONS:
- With undiluted acid:
At 0.25 ml/kg, rats were sluggish and presented an unsteady gait.
At 0.5 ml/kg, they exhibited the same symptoms as above. Death occured within 40 minutes.
At 1.0 ml/kg, sluggish and unsteady gait were noted. Deep breathing was immediately observed.

- With potassium salt (0.25 g/ml):
At 2 g/kg: no symptoms
At 4 g/kg: sluggish behavior
At 8 g/kg: sluggish gait and pilo-erection.

MORTALITY:
- With undiluted acid:
At 0.25 ml/kg, 2 rats (among 5) died within 40 minutes.
At 0.5 ml/kg, all rats died (2 on first day, 2 on second day and 1 on fifth day)
At 1.0 ml/kg, 2 rats (among 3) died within 45 minutes.

- With potassium salt (0.25 g/ml):
At 2 g/kg: no death
At 4 g/kg: no death
At 8 g/kg: 4 rats (among 5) died: 3 on first day and 1 on second day.

NECROPSY:
- With undiluted acid:

- With undiluted acid:
Livers mottled and burned, stomach burned, pylorus hemorrhaged and gas filled, intestines hemorrhaged, injected and gas filled, kidneys mottled and slightly congested.

- With potassium salt (0.25 g/ml):
Livers and spleens mottled, kidneys speckled and slightly congested, stomachs and intestines distented and liquid filled.

LD50:
- With undiluted acid:
0.281 ml/kg (0.146-0.540)

- With potassium salt (0.25 g/ml):
6.17 g/kg (4.57-8.33)

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Harlan Industries, Cumberland.
- Age: 30 days
- Weight at study initiation: 84 to 147 g (males) and 79 to 130 g (females)
- Number of animals: 5 rats / dose (except 3 rats / lower dose)
- Controls: no
- Other:

ADMINISTRATION:
- Exposure route: gavage
- Volume administered: no data
- Post dose observation period: 5 days (all rats were dead on the fifth day)

EXAMINATIONS: clinical observations, mortality and necropsy

Test substance : Source: South Charleston, WV.
Batch number: 8E29
Purity: 98%
Other: Methane sulfonic acid and its potassium salt were tested

Conclusion : Sodium salt was found to be 20 times less toxic than undiluted acid.

Reliability : (2) valid with restrictions

18.12.2002 (29)

Type : LD50

Value : ca. 200 - 400 mg/kg bw

Species : rat

Strain :

Sex :

Number of animals :

Vehicle :

Doses :

Method : other

Year :

GLP : no

Test substance : as prescribed by 1.1 - 1.4

Remark : Study peer reviewed

Source : ATOFINA, Paris-La Défense, France.

Reliability : (4) not assignable
documentation insufficient for assessment.

18.12.2002 (30)

5.1.2 ACUTE INHALATION TOXICITY

Type	:	LC0
Value	:	
Species	:	mouse
Strain	:	no data
Sex	:	male
Number of animals	:	5
Vehicle	:	no data
Doses	:	saturated atmosphere
Exposure time	:	1 hour(s)
Method	:	other
Year	:	1976
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	No effects of any kind were discernible in any of the twenty mice either during or after exposure for 1 hour to saturated vapor (at 20°C). All showed normal gains in body weight during the subsequent 7-day observation period.
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	Source: no data Batch number: no data Purity: no data
Test substance	:	TEST ORGANISMS: - Source: no data - Age: no data - Weight at study initiation: 32 g - Number of animals: (per sex and dose) - Controls: no ADMINISTRATION: - Type of exposure: whole body - Other: Atmospheric saturation was obtained by placing 2 g of the sample in an airtight twenty-litre chamber and allowing 24h for evaporation. The chamber was then momentarily opened to permit the quick insertion of mice. EXAMINATIONS: clinical observations and body weight.
Conclusion	:	Insufficient volatility of methane sulfonic acid limits the actual exposure and therefore toxicity risks.
Reliability	:	(2) valid with restrictions Lack of information about the method.
Flag	:	Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

30.12.2002 (31)

Type	:	LC0
Value	:	
Species	:	rat
Strain	:	no data
Sex	:	no data
Number of animals	:	
Vehicle	:	
Doses	:	
Exposure time	:	6 hour(s)
Method	:	other
Year	:	
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	No effect in rats after 6 hours of inhaling vapor from heated liquid. Study peer reviewed

Study peer reviewed
Source : ATOFINA, Paris-La Défense, France.
Reliability : (4) not assignable
documentation insufficient for assessment.

27.12.2002 (32)

5.1.3 ACUTE DERMAL TOXICITY

Type : LD0
Value : > 1000 mg/kg bw
Species : rabbit
Strain : New Zealand white
Sex : male/female
Number of animals : 10
Vehicle : water
Doses : 1000 mg/kg
Method : OECD Guide-line 402 "Acute dermal Toxicity"
Year :
GLP : yes
Test substance : other TS

Method : Initially, one healthy male New Zealand White rabbit was dosed dermally at 1000 mg/kg of Methane Sulfonic Acid (MSA) (test sample of 70% MSA in water was diluted to yield 200 mg MSA/ml, dose volume was 5 mg/kg). Per the sponsor's request, an additional nine animals (4 males and 5 females) were dosed. The test article was kept in contact with the skin for 24 hours. Dermal responses were recorded 24 hours postdose and on days 7 and 14. Animals were observed for mortality, toxicity and pharmacological effects at 1 and 2 hours postdose and once daily for 14 days. Body weights were recorded pretest and at termination. All animals were examined for gross pathology.

Result : All animals survived the dermal application of 1000 mg/kg Methane Sulfonic Acid.

Instances of few faces were the only abnormal physical signs noted during the observation period.

Dermal responses were slight to well defined on day 1, absent to severe on day 7 and absent to slight on day 14.
Body weight changes were normal.

Necropsy results were normal.

Source : Atofina, Paris-La-Defense, France
Test substance : Origine: Atofina Chemicals
Batch: 20330PI
Purity: 70.2% in water
Conclusion : The LD50 of Methane Sulfonic Acid is greater than 1000 mg/kg of body weight.
Reliability : (1) valid without restriction
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

30.12.2002 (33)

Type : LD50
Value : ca. 200 - 2000 mg/kg bw
Species : rabbit
Strain : other: albino
Sex :
Number of animals : 6
Vehicle : other: undiluted (2000 mg/kg) and water (200 mg/kg)

Doses	:	200 and 2000 mg/kg
Method	:	other
Year	:	1978
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	At 2000 mg/kg: Skin contact caused intense and prolonged pain. At the end of exposure, the skin of each animal was dark grey in appearance and scattered portions were separating from the subcutaneous tissues. 2 rabbits died the following night (28h after initial contact) and 1 rabbit was euthanized three days after treatment for humane reasons. At 200 mg/kg : No mortality. Erythema (score 2) was present over the entire trunk of each animal together with numerous small lesions which resembled acid burns. Animals nevertheless remained asymptomatic and gained body weight during the observation period.
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	TEST ORGANISMS: - Source: no data - Age: no data - Weight at study initiation: no data - Number of animals: no data - Controls: no ADMINISTRATION: - Area covered: no data - Exposure: pre-fitted impervious sleeve - Concentration in vehicle: undiluted or 10% (w/v) - Total volume applied: no data - Removal of test substance: with a saturated solution of sodium bicarbonate - Exposure duration: 24h - Post-dose observation period: 7 days EXAMINATIONS: clinical observations and mortality
Test substance	:	Source: no data Batch number: no data Purity: no data
Conclusion	:	A 2000 mg/kg dermal exposure to methane sulfonic acid is highly toxic (severe skin injuries causing death) whereas 200 mg/kg exposure induces minor skin lesions (erythema and acid burns) but no systemic toxicity.
Reliability	:	(2) valid with restrictions Details on the protocol used are not available.

03.01.2003 (34)

Type	:	LD50
Value	:	> 2000 mg/kg bw
Species	:	guinea pig
Strain	:	
Sex	:	
Number of animals	:	
Vehicle	:	
Doses	:	
Method	:	other: no data
Year	:	
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4

Remark	:	LD50 > 20 ml (of the 10% solution in water) Study peer reviewed
Source	:	Atofina, Paris-La Défense, France.
Test substance	:	10% MSA in water.
Reliability	:	(4) not assignable Documentation insufficient for assessment.

03.01.2003 (32)

5.1.4 ACUTE TOXICITY, OTHER ROUTES

Type	:	LC50
Value	:	< 50 mg/kg bw
Species	:	rat
Strain	:	
Sex	:	
Number of animals	:	
Vehicle	:	
Doses	:	
Route of admin.	:	i.p.
Exposure time	:	
Method	:	no data
Year	:	
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Source	:	ATOFINA, Paris-La Défense, France.
Reliability	:	(4) not assignable Documentation insufficient for assessment.

18.12.2002 (32)

5.2.1 SKIN IRRITATION

Species	:	mouse
Concentration	:	
Exposure	:	
Exposure time	:	
Number of animals	:	
Vehicle	:	
PDII	:	
Result	:	corrosive
Classification	:	corrosive (causes burns)
Method	:	other: Mouse tail method
Year	:	1976
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	The tail of each mouse was white when exposure was terminated and the appendage fell off in a day or two.
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	The tails of four anesthetized mice were immersed in MSA to a depth of 4 cm. One hour later the tissue reaction was terminated by plunging the appendages into a saturated solution of sodium bicarbonate for a period of 30 seconds.
Conclusion	:	Methane sulfonic acid is corrosive to mouse skin
Reliability	:	(2) valid with restrictions Lack of information about the "Mouse tail method".

Flag	:	Material Safety Dataset, Directive 67/548/EEC
18.12.2002		(31)

5.2.2 EYE IRRITATION

Species	:	rabbit
Concentration	:	
Dose	:	.1 ml
Exposure time	:	
Comment	:	other: see Test Conditions freetext about rinse
Number of animals	:	2
Vehicle	:	none
Result	:	corrosive
Classification	:	risk of serious damage to eyes
Method	:	other
Year	:	1978
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	Each instillation caused excruciating pain. Unwashed eye: The reaction involved all ocular tissues and occured immediately. The conjunctivae became completely necrotic (white) without evident swelling. The iris was dilated with ragged edges and failed to react to light. The cornea opacified completely within 24 hours. Washed eye: The reaction did not differ significantly from that of unwashed eye.
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	TEST ORGANISMS: - Strain: Albino - Sex: no data - Source: no data - Age: no data - Weight at study initiation: no data - Controls: auto-control ADMINISTRATION: - Administration frequency: single administration - Other: one of the two animals had a rinse of the treated eye with flowing water (initiated 20 to 30 seconds after instillation and continued for one minute). EXAMINATIONS: - Ophthalmoscopic examination: Yes - Scoring system: not mentioned - Observation period: 10 min, 1h, 2h, 3h, 4h, 24h, 48h, 72h, 4 days, 5 days, 6 days and 7 days. - Tool used to assess score: no data
Test substance	:	Source: no data Batch number: no data Purity: no data
Conclusion	:	Methane sulfonic acid is extremely corrosive to eye.
Reliability	:	(2) valid with restrictions Even if method used differs from guidelines, corrosivity of methane sulfonic acid is evidently highlighted and does not require additional animal testing.
Flag	:	Material Safety Dataset, Directive 67/548/EEC

18.12.2002 (35)

5.3 SENSITIZATION

Type	:	other: Buehler derived test
Species	:	guinea pig
Concentration	:	1st: Induction 50 % occlusive epicutaneous 2nd: Challenge 25 % occlusive epicutaneous 3rd:
Number of animals	:	30
Vehicle	:	water
Result	:	not sensitizing
Classification	:	not sensitizing
Method	:	other: not mentionned
Year	:	
GLP	:	yes
Test substance	:	other TS
Remark	:	Study peer reviewed
Result	:	RESULTS OF PILOT STUDY: Undiluted methane sulfonic acid 70% produced grades of 2, 1 and ±, with oedema, blanching and scabbing on two sites. 50%, 25%, 10%, 5%, 2.5% and 1% produced grades of ±, while 0.5% produced grades of ± and 0. Consequently, the 50% concentration was chosen for use at induction for the test group, since it caused no greater than mild to moderate primary irritation. Moreover, the 25% concentration was selected for challenge, because it caused no more than slight irritation. RESULTS OF TEST - Sensitization reaction: None of the test animals responded with a skin grade that would have been suggestive of sensitization. - Clinical signs: no data
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	TEST ORGANISMS: - Strain: Hartley - Sex: male and female - Source: Harlan Sprague Dawley (P.O. Box 29176, Indianapolis, Indiana 46229) - Age: no data - Weight at study initiation: 429.6 ± 18.8 (males), 400.5 ± 27.6g (females) - Number of animals: 10 males + 10 females - Controls: 5 males + 5 females ADMINISTRATION: - Induction schedule: - On day 1, dermal application with 0.3 ml of test substance (treated group) or with the vehicle (control group) on the left shoulder. - On day 7, the same region received another topical application - On day 14, this same site was treated by a last topical application All these applications lasted approximately 6 hours. - Concentration in Freunds Complete Adjuvant (FCA): not used here - Challenge schedule: On day 28, all the animals received 0.3 ml of the test substance at the concentration of 25% in their right flank. - Challenge exposure duration : 24h - Rechallenge: no - Positive control: no - Other: EXAMINATIONS: - Examination schedule: 24h after each induction an challenge application.

- Examination schedule: 24h after each induction an challenge application.
- Grading system:
0: no reaction
±: slight, patchy erythema
1: slight but confluent or moderate patchy erythema
2: moderate erythema
3: severe erythema with or without oedema.

- Pilot study: Yes
A preliminary study was conducted in order to determine the concentrations to be tested in the main study.
The irritation potential of methane sulfonic acid 70% at levels of undiluted, 50%, 25%, 10%, 5%, 2.5%, 1% and 0.5% was evaluated in two groups of four animals each. Four levels of test material were evaluated per animals.
Dilutions were obtained with distilled water (w/v).
0.3 ml of different solutions was applied into a 25 mm Hill Top Chamber, which were placed on animals clipped back for 6 hours. The day after, animals were depilated and two hours later, examined for irritation, according the previously described scale.
Another score was performed 40h after exposure.

Test substance : Source: Elf Atochem
Batch number: M12E
Purity: no data

Conclusion : 20 guinea-pigs were induced by 50% of methane sulfonic acid 70% solution (i.e. 35% of methane sulfonic acid) and challenged by 25% of methane sulfonic acid 70% solution (i.e. 17.5% of methane sulfonic acid). None of the test animals responded with a skin grade that would have been suggestive of sensitization.

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC

18.12.2002 (36)

5.4 REPEATED DOSE TOXICITY

Type :
Species : rat
Sex : male/female
Strain : other: Albino
Route of admin. : inhalation: aerosol
Exposure period : 4 weeks
Frequency of treatm. : 6 hours/day, 5 days/week
Post exposure period : 2-week recovery period
Doses : 0.026, 0.073 and 0.242 mg/L
Control group : yes, concurrent vehicle
NOAEL : = .026 mg/l
Method : other: EPA 40 CFR 792
Year :
GLP : yes
Test substance : other TS

Method : - Analytical device:
Ion exchange chromatography, Intox Products particle size analyzer model 02-140/JB, Teflon 25-mm filter and Mettler AE240 electronic balance (for gravimetric determination).
- Statistical tests:
One-way analysis of variance followed by a Dunnett's test using a Digital MicroVAX 3400 computer "with appropriate programming".
Fisher's Exact test was involved for discontinuous (ordinal or descriptive) functional observation battery data.
Clinical laboratory values for leukocytes that occur at a low incidence

(monocytes, eosinophils, basophils, unsegmented neutrophils) were not subjected to statistical analysis.

Remark : Study peer reviewed

Result : - Mortality:
During the exposure phase of the study, 4, 1, 1 and 5 animals in the control, low-, mid- and high-exposure groups, respectively, were found dead. All deaths were attributed to confinement in the restraint tubes and none of the deaths were considered related to exposure to the test article. All other animals survived to the scheduled necropsies.

- Clinical signs:
Exposure-related clinical signs consisted of rales in the high-exposure group animals and an increased incidence of yellow matting on various body surfaces in the mid-exposure group males and the high-exposure group males and females. No significant clinical findings were noted in any animals during the recovery period.

- Body weight gain:
A transient reduction in mean body weight gain of high-exposed males during the first week of exposure.

- Food consumption:
The high-exposure group males experienced slightly decreased food consumption means throughout the exposure period.

- Haematology:
No adverse effects

- Biochemistry:
No adverse effects
At week 4 evaluation, a statistically significant increase was noted in blood urea nitrogen and aspartate-amino-transferase in the high-exposure group males and females, respectively. Such increases were not considered to be of toxicological significance since blood urea nitrogen and aspartate-amino-transferase were comparable to control values after the two-week recovery period.

- Urinanalysis:
No adverse effects

- Organ weights:
No significant changes were noted.

- Gross pathology:
Gross findings in the animals that were found dead during the study (dark red content of ileum, dark red lungs, reddened and/or enlarged lymph nodes, etc.) were seen at a similar incidence in the control group or are not uncommon in rats.

- Histopathology:
Test article-related microscopic findings were observed in the nasal turbinates of the high-exposure group rats that were found dead and in all treated groups at the study week 4 and 6 evaluations; however, the severity of some of the findings observed after the two-week recovery period suggested at least partial recovery from the irritative effects of the test article.

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Charles River Breeding Laboratories (Portage, Michigan)
- Age: 36 to 43 days
- Weight at study initiation: 235 ± 29.4 g for males, 188 ± 16.9 g for females
- Number of animals: 120 rats : 15 males + 15 females / dose group (3 dose groups + 1 control group)

dose groups + 1 control group)

ADMINISTRATION:
- Type of inhalation study: nose only
- Particle size:
. MMAD (Mass Median aerodynamic Diameter): 1.1
. Geometric Standard Deviation: 1.8
- Type or preparation of particles: Inspiron Model 002305-A nebulizer (Inertech Resources)
- Vehicle: filtered air
- Nominal / analytical concentrations:
0.025 / 0.026 mg/l
0.075 / 0.073 mg/l
0.25 / 0.242 mg/l

SATELLITE GROUPS AND REASONS THEY WERE ADDED: none

CLINICAL OBSERVATIONS AND FREQUENCY:
- Clinical signs: yes (twice a day on exposure days and once a day on non-exposure days)
- Mortality: yes (twice a day)
- Body weight: yes (one week prior initiation of test article exposure [day-7] and prior to necropsy).
- Food consumption: yes (one week before the beginning of exposure, then weekly)
- Ophthalmoscopic examination: no
- Haematology: yes
Hemoglobin, hematocrit, red blood cell count, white blood cell count, differential leukocyte count, platelet count, mean cell volume, mean cell haemoglobin concentration, mean cell haemoglobin
- Biochemistry: yes
- Electrolytes: calcium, chloride, phosphorous, potassium, sodium,
- Enzymes: alkaline phosphatase, alanine-aminotransferase, aspartate-aminotransferase, gamma-glutamyl-transferase
- Other: albumin, blood creatinine, blood urea nitrogen, albumin/globulin, glucose, total bilirubin, total cholesterol, total serum protein, bile acids
- Urinanalysis: yes
urine volume, pH, specific gravity, proteins, glucose, ketones, bilirubin, nitrites, blood, urobilinogen, leukocytes, microscopy of sediments, appearance, color.

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Weighted organs: adrenals, brain, heart, kidneys, liver, lungs, ovaries, spleen, testes with epididymides and thymus.
- Macroscopic examined
- Cardio-vascular and hematopoietic system: heart, aorta (thoracic), bone marrow, lymph node (bronchial, mesenteric, suprapharyngeal), thymus, spleen
- Digestive system: tongue, salivary glands (maxillary), oesophagus, stomach, liver, gallbladder, pancreas, duodenum, jejunum, ileum, caecum, colon, rectum
- Glandular system: pituitary, thyroid, parathyroids, adrenal, mammary gland
- Nervous system: brain, spinal cord, sciatic nerve, eye (optic nerve)
- Respiratory system: nasal turbinates, trachea, lungs with bronchi
- Uro-genital system: kidney, bladder, ovary, uterus, vagina, testes, epididymides, seminal vesicle, prostate, Fallopian tubes
- Other: skin, muscle (vastus medialis)
- Microscopic: lungs and nasal cavity
- Cardio-vascular and hematopoietic system: heart, aorta (thoracic), bone marrow, lymph node (bronchial, mesenteric, suprapharyngeal), thymus, spleen

spleen
- Digestive system: tongue, salivary glands (maxillary), oesophagus, stomach, liver, gallbladder, pancreas, duodenum, jejunum, ileum, caecum, colon, rectum
- Glandular system: pituitary, thyroid, parathyroids, adrenal, mammary gland
- Nervous system: brain, spinal cord, sciatic nerve, eye (optic nerve)
- Respiratory system: nasal turbinates, trachea, lungs with bronchi
- Uro-genital system: kidney, bladder, ovary, uterus, vagina, testes, epididymides, seminal vesicle, prostate, Fallopian tubes
Other: skin, muscle (vastus medialis)

OTHER EXAMINATIONS: none

Test substance : Source: Elf Atochem
Batch number: M-12-E
Purity: 69.9%
Other: substance was supplied in a 70% aqueous solution

Conclusion : Exposure of albino rats to an aerosol of methane sulfonic acid resulted in changes in the clinical conditions of the animals at exposure levels of 0.073 and 0.242 mg/L, a transient reduced mean body weight gain in the 0.242 mg/L exposure level males, slightly reduced food consumption means in the 0.242 mg/L exposure level males and histopathologic lesions in the nasal turbinates of all exposed groups.
Based on the data obtained during the two-week recovery (nonexposure) period, the effects on the clinical conditions of the animals and body weight gains were completely reversible; histopathological lesions in the nasal turbinates were observed in all groups at the end of the recovery period.
Based on the compound-induced lesions observed in the nas al turbinates, the no observed effect level (NOEL) for local irritation was considered to be less than 0.026 mg/L. The NOEL for systemic toxicity was considered to be 0.026 mg/L.

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

27.12.2002 (37)

Type :
Species : rat
Sex : male/female
Strain : Sprague-Dawley
Route of admin. : inhalation
Exposure period : 5 days
Frequency of treatm. : 6 hours/day
Post exposure period : no
Doses : 0.026, 0.082, 0.23 and 0.74 mg/L
Control group : yes, concurrent vehicle
NOAEL : = .082 mg/l
Method : other: EPA 40 CFR 792
Year :
GLP : yes
Test substance : other TS

Method : - Analytical device:
Ion chromatography, Intox Products particle size analyzer model 02-140/JB, Intox Products 25-mm filter and Mettler AE240 electronic balance (for gravimetric determination).
- Statistical tests:
One-way analysis of variance followed by a Dunnett's test using a Digital MicroVAX 3400 computer "with appropriate programming".

Remark : Study peer reviewed

Result : - Mortality:
Two and three test article-related deaths occurred in the 0.23 and 0.74 mg/L groups, respectively. All other animals survived to the scheduled

mg/L groups, respectively. All other animals survived to the scheduled necropsy.

- Clinical signs:
The predominant test article-related clinical sign in males and females in the 0.23 and 0.74 mg/L groups was rales.

- Body weight gain:
Mean body weight losses and/or reduced mean body weight gains were noted in males and females in the 0.74 mg/L group during days 0-1 through 2-3; a slight mean body weight loss occurred in males in this group during day 4-5. Mean body weights were generally reduced in males and females in this group throughout the study.

- Food consumption:
Food consumption was generally reduced throughout the study relative to the control group values in males and females in the 0.74 mg/L group.

- Organ weights:
Organ weights were unaffected by test article administration at any exposure level.

- Gross pathology:
No treatment-related macroscopic findings were noted in treated animals

- Histopathology:
Microscopic findings in the nasal cavities that were considered to be related to exposure to the test article included mucosal necrosis, suppurative inflammation and/or nasal exudate in males and females in the 0.23 and 0.74 mg/L groups.

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Charles River Breeding Laboratories (Portage, Michigan)
- Age: 36 days
- Weight at study initiation: 214 ± 16 g for males, 159 ± 11.7 g for females
- Number of animals: 50 rats : 5 males + 5 females / dose group (4 dose groups + 1 control group)

ADMINISTRATION:
- Type of inhalation study: nose only
- Particle size: see corresponding attached document
- Type or preparation of particles: De Vilbis glass nebulizer
- Vehicle: air
- Nominal / analytical concentrations:
0.022 / 0.0259 mg/l
0.066 / 0.0818 mg/l
0.22 / 0.2264 mg/l
0.66 / 0.7387 mg/l

SATELLITE GROUPS AND REASONS THEY WERE ADDED: none

CLINICAL OBSERVATIONS AND FREQUENCY:
- Clinical signs: yes (twice a day)
- Mortality: yes (twice a day)
- Body weight: yes (one week prior initiation of test article exposure [day-7], once before the allocation of animals to groups [day-1], every day of treatment and then on day 5, prior to necropsy).
- Food consumption: yes (daily, one week before the beginning of exposure)
- Ophthalmoscopic examination: no
- Haematology: no
- Biochemistry: no

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Macroscopic examined and weighted organs: external surface, all orifices, cranial, thoracic, abdominal and pelvic cavities including viscera, with particular attention paid to the nasal and lung tissues. A the time of necropsy, gross lesions, heads and lungs were collected and placed in a 10% neutral buffered formalin.
- Microscopic: lungs and nasal cavity

OTHER EXAMINATIONS: none

Test substance : Source: Elf Atochem
Batch number: L-04-D
Purity: 70%
Other: substance was supplied in a 70% aqueous solution

Attached document : 75-75-25-day inhalation test - Particle size.bmp

Analytical concentrations (mg/L)	MMAD (µm)	AGSD
0,0269	1,4	1,7
0,0818	1,4	1,9
0,2264	1,4	1,8
0,7387	1,5	1,9

MMAD: Mean Median Average Diameter
AGSD: Average Geometric Standard Deviation

Conclusion : Methane sulfonic acid, administered via nose-only inhalation for six hours per day for five consecutive days, caused mortalities, clinical signs of toxicity and microscopic effects on nasal cavities at exposure levels of 0.23 and 0.74 mg/L and inhibition of body weight gain and food consumption at an exposure level of 0.74 mg/L.
No signs of systemic toxicity were observed at exposure levels of 0.026 and 0.082 mg/L.
The NOEL for systemic toxicity was found to be 0.082 mg/L.
Based on data obtained, exposure level of 0.025, 0.075 and 0.25 mg/L were chosen for a subsequent 28-day inhalation study.

Reliability : (1) valid without restriction
18.12.2002 (38)

Type :
Species : rat
Sex : male/female
Strain : Wistar
Route of admin. : oral feed
Exposure period : 7 days
Frequency of treatm. : ad libitum
Post exposure period : none
Doses : Males: 0, 51, 185, 420, 1805 mg/kg bw/d; Females: 0, 55, 201, 551, 2122 mg/kg bw/d
Control group : yes, concurrent no treatment
NOAEL : >= 1805 - 2122 mg/kg bw
Method : other
Year : 1975

GLP : no
Test substance : other TS

Method : Statistical tests:
For growth effects the means for each sex were calculated and compared after adjustment of the individual weights of each rat to a change in weight. over their weight on the morning of their first day of dosing. The results of weight changes were intercompared for the dosage groups by use of Bartlett's test for homogeneity of variance, by the analysis of variance and by Duncan's multiple range test. The latter was used, if F for the analysis of variance was significantly high, to delineate which groups differed from the other. If Bartlett's test indicated heterogeneous variances, the F-test was used for any paired-group comparison. If these individual F-tests were not significant, Student's t-test was used; if significant, the means were compared by the Cochran t-test. The fiducial limit of 0.05 was employed as the critical level of difference not believed to be produced by chance.

Remark : Study peer reviewed

Result : Mortality: no death occured
Body weight: no significant variation
Organ weights: no relevant changes in terms of doses

Detailed data are presented in the attached document.

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Harlan Industries, Cumberland.
- Age: 30 days
- Weight at study initiation: 84 to 147 g (males) and 79 to 130 g (females)
- Number of animals: 5 rats / dose (except 3 rats / lower dose)
- Controls: yes

ADMINISTRATION:
- Vehicle: distilled water then mixed with food
- Concentration in diet: 0.043%, 0.159%, 0.382% and 1.635% (for males) and 0.045%, 0.183%, 0.479% and 1.80% (for females)
- Food consumption: 13.9 to 16.9 g of food/day
- Other: After the first day of dose, the dosage level of the rats on 10 mg/kg of methane sulfonic acid in diets was increased to 2000 mg/kg/day for the reminder of the week.

SATELLITE GROUPS AND REASONS THEY WERE ADDED: none

CLINICAL OBSERVATIONS AND FREQUENCY:
- Clinical signs: yes (at least once a day)
- Mortality: yes (at least twice a day)
- Body weight: yes (three times during the week)
- Food consumption: yes (no data about frequency)
- Water consumption: no data
- Ophthalmoscopic examination: no
- Haematology: no
- Biochemistry: no
- Urinalysis: no

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Macroscopic examined and weighted organs: liver and kidneys
- Microscopic: none

Test substance : Source: South Charleston, WV.
Batch number: 8E29
Purity: 98%
Other: Methane sulfonic acid and its potassium salt were tested

Attached document : 75-75-2 Sub-acute EPA-OTS0536043.bmp

Ref. 38-1

Summary of Results of 7 Days of Inclusion of Methanesulfonic Acid and its Potassium Salt in the Diet of Rats

	Methane Sulfonic Acid (MSA)					Potassium Salt of M	
	Male Rats						
Dosage goal, mg/kg	2000	500	200	50	0	2000	0
Concentration in diet, % x 10^{-3}	1635	382	159	43	0	1712	0
Dosage attained, mg/kg/day	1805	420	185	51	0	2080	0
Diet consumed, gm/rat/day	16.3	14.2	15.9	16.9	15.9	16.4	15.9
Body weight change, gm							
1 day of doses	-x	2.2	5.2	3.6	2.8	6.8	6.4
4 days of doses	28.2	20.6	27.2	25.4	24.2	25.4	26.2
7 days of doses	45.0	34.6	41.0	39.8	38.4	47.6	47.2
Liver weight, gms	8.02	6.51	7.07	7.68	7.20	7.26	7.1
Liver wt. as % of body wt.	4.70	4.39	4.48	4.72	4.72	4.58	4.5
Kidney weight, gms	1.64	1.46	1.48	1.55	1.44	1.54	1.4
Kidney wt. as % of body wt.	0.96	0.98	0.94	0.96	0.95	0.97	0.9
Mortality	0	0	0	0	0	0	0
	Female Rats						
Dosage goal, mg/kg	2000	500	200	50	0	2000	0
Concentration in diet, % x 10^{-3}	1800	479	183	45	0	1657	0
Dosage attained, mg/kg/day	2122	551	201	55	0	1960	0
Diet consumed, gm/rat/day	14.7	15.4	13.9	15.2	15.5	13.6	14.3
Body weight change, gms							
1 day of doses	-x	5.0	4.0	3.8	3.8	5.2	5.2
4 days of doses	18.6	21.4	18.6	20.6	19.0	20.4	21.6
7 days of doses	33.4	36.8	32.0	35.0	37.4	38.2	34.4
Liver weight, gms	6.68	7.36	6.76	6.86	7.56	6.22	6.2
Liver wt as % of body wt	4.70	4.84	4.71	4.78	5.18	4.63	4.7
Kidney weight, gms	1.33	1.41	1.30	1.34	1.41	1.24	1.2
Kidney wt as % of body wt	0.94	0.93	0.92	0.94	0.97	0.93	0.9
Mortality	0	0	0	0	0	0	0

x Rats were fed 10 mg/kg for one day and then fed 2000 mg/kg for the subsequent 6 days

Conclusion : None of the rats orally exposed to methane sulfonic acid (up to 1800 mg/kg/day) or to its potassium salt (2000 mg/kg/day) died during this 7-day study.
Furthermore, none of the measured parameters (food consumption, body weight change, liver and kidney weight) was affected by the exposure.
Consequently, NOAEL can be valued at 1805 mg/kg/day for males and 2122 mg/kg/day for females.

Reliability : (2) valid with restrictions
18.12.2002 (29)

5.5 GENETIC TOXICITY 'IN VITRO'

Type : Ames test
System of testing : TA1535, TA1537, TA1538, TA98 and TA100
Test concentration : 100, 500, 1000, 2500 and 5000 µg/plate
Cycotoxic concentr. : 5000 and 10000 µg/plate on TA98 and TA100
Metabolic activation : with and without
Result : negative
Method : OECD Guide-line 471
Year : 1983
GLP : yes
Test substance : other TS

Method : Analytical device: Artek 980 electronic colony counter
Statistical test: no data
Remark : Study peer reviewed
Result : BACTERIAL TOXICITY
Methane sulfonic acid induced a marked toxicity on two tester strains: TA98 and TA100 at 1000 µg/plate with and without metabolic activation. In addition it was observed that the S9 mix preparation precipitated at those two concentrations.
Without metabolic activation at 5000 µg/plate, a moderate toxic effect was noted on TA100, whereas on TA98 a decrease of the revertant colonies number with a sparse background was noted.
Therefore, the following concentrations were selected for the genotoxicity

Therefore, the following concentrations were selected for the genotoxicity study: 100, 500, 1000, 2500 and 5000 µg/plate.

GENOTOXIC EFFECTS:
Whatever conditions used (metabolic activation or not), methanesulfonic acid did not induce any increase of revertant colonies per plate, at any dosages. This is true for both assays.

PRECIPITATION:
A precipitation of the S9 mix preparation was observed from the concentration of 2500 µg/plate, in a concentration-related effect of increasing intensity in the first assay, while it only appeared at 5000 µg/plate in the second one.

TEST-SPECIFIC CONFOUNDING FACTORS: none

Source : ATOFINA, Paris-La Défense, France.

Test condition : EXPERIMENTAL CONDITIONS:
- Number of replicates: 3/dose
- Metabolic activation: S9-mix (from male Sprague-Dawley rats, treated by Aroclor 1254)
- Vehicle: distilled water
- Negative control: distilled water
- Positive controls:
. for TA1535 and TA100: sodium azide (5µg/plate)
. for TA1538 and TA98: 2-nitrofluorene (5µg/plate)
. for TA1537: 9 -aminoacridine (100µg/plate)
. for all strains: 2-aminoanthracene (5µg/plate except 2.5µg/plate for TA1537)
- Pre-incubation time: 20 minutes
- Pre-incubation temperature: 37°C
- Incubation time: 48 hours
- Incubation temperature: 37°C

EXAMINATION:
- Bacterial toxicity (performed on TA100 and TA98 at 500, 1000, 5000 and 10000 µg/plate)
- Number of revertants / plate

CRITERIA FOR EVALUATING RESULTS:
- Positivity criteria for cytotoxicity:
. reduced numbers of revertant colonies/plate compared with control plates
. sparsity of bacterial background lawn when compared with control plates
- Positivity criteria:
. number of revertants at least twice that of spontaneous revertants
. dose-related pattern
. number of revertant/nmol > 0.01
. reproducibility of the positive response

Test substance : Source: Penwalt
Batch number: ML155
Purity: 98.8%

Conclusion : No genotoxicity was observed on tested strains (TA1535, TA1537, TA1538, TA98 and TA100) up to the cytotoxicity threshold (5000 µg/plate).

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Critical study for SIDS endpoint

27.12.2002 (39)

Type : Ames test

System of testing : Salmonella typhimurium TA98, TA100, TA1535, TA1537, TA1538

Test concentration : 10, 32, 100, 316, 1000 µg/plate

Cycotoxic concentr. : 1000 µg/plate

Metabolic activation : with and without

Result : negative

Method	:	OECD Guide-line 471
Year	:	1983
GLP	:	yes
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	No increases in reversion to prototrophy were obtained with any of the five bacterial strains at the compound level tested, either in the presence or absence of S9 mix. Inhibition of growth, observed as thinning of the background lawn of non revertants cells and reduction in colony numbers, occured in all strains following exposure to 1000 µg/plate.
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	EXPERIMENTAL CONDITIONS: - Number of replicates: no data - Metabolic activation: S9-mix - Vehicle: no data - Negative control: water - Positive controls: sodium azide, 2-nitrofluorene, 9-aminoacridine, 2-aminoanthracene and benzo(a)pyrene - Pre-incubation time: no data - Pre-incubation temperature: no data - Incubation time: no data - Incubation temperature: no data EXAMINATION: - Growth inhibition - Number of revertants / plate CRITERIA FOR EVALUATING RESULTS: no data
Test substance	:	Source: no data Batch number: no data Purity: no data
Conclusion	:	Methane sulfonic acid is devoid of mutagenic activity.
Reliability	:	(1) valid without restriction
Flag	:	Material Safety Dataset, Critical study for SIDS endpoint

03.01.2003 (40)

Type	:	Ames test
System of testing	:	Salmonella typhimurium TA100, TA1535
Test concentration	:	384.4 to 12300 µg/plate
Cycotoxic concentr.	:	no data
Metabolic activation	:	with and without
Result	:	negative
Method	:	other: Mutation Res., 113: 173-215, 1983
Year	:	1989
GLP	:	no
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	Study peer reviewed
Result	:	see attached document
Source	:	ATOFINA, Paris-La Défense, France.
Test condition	:	EXPERIMENTAL CONDITIONS: - Number of replicates: 3/dose - Metabolic activation: S9-mix (from male Sprague-Dawley rats, treated by Aroclor 1254) - Vehicle: distilled water - Negative control: distilled water - Positive control: benzo(a)pyrene (S9+) and diepoxybutane (S9-) - Pre-incubation: no - Incubation time: no data

- Incubation time: no data
- Incubation temperature: no data

EXAMINATION:
- Bacterial toxicity
- Number of revertants / plate. The test was repeated.

Test substance : Source: Aldrich
Batch number: no data
Purity: 99%

Attached document : 75-75-2 Ames test (Zeiger).bmp

TABLE II. Mutagenicity of Methanesulfonic Acid in *Salmonella* Strains TA1535 and TA100 Without (A) and With (B) S9, in the Plate Test[a]

	TA1535		TA100	
μmol/plate	His⁺/plate	Ind. Revs.[b]	His⁺/plate	Ind. Revs.
(A) - S9				
0	9 ± 2	0	130 ± 15	0
4	15 ± 6	6	120 ± 11	−10
8	16 ± 6	7	117 ± 13	−13
16	15 ± 3	6	119 ± 21	−11
32	15 ± 8	6	136 ± 15	6
64	17 ± 4	8	143 ± 21	13
128	13 ± 2	4	132 ± 5	2
DEB (1)[c]	125 ± 6	116	390 ± 16	260
(B) + S9				
0	16 ± 0	0	127 ± 3	0
4	16 ± 3	0	132 ± 6	5
8	15 ± 4	−1	143 ± 14	16
16	18 ± 5	2	146 ± 22	19
32	18 ± 5	2	132 ± 12	5
64	16 ± 5	0	153 ± 7	26
128	19 ± 3	3	144 ± 8	17
BaP (0.02)	–[d]		1787 ± 106	1660

[a]His⁺ revertants/plate (counts from triplicate plates) ± S.D.
[b]Induced his⁺ revertants (spontaneous control subtracted).
[c]Positive control (dose): DEB, dl-1,2:3,4-diepoxybutane; BaP, benzo(a)pyrene.
[d]Not done.

Reliability : (2) valid with restrictions
Only 2 strains tested and scarce data.

03.01.2003 (41)

Type : other: Ames-derived gradient technique
System of testing : E. coli (WP2 and WP2 uvrA-), S. typhimurium (G46, TA1535, TA100, C3076, TA1537, D3052, TA1538, TA98)
Test concentration : No data available
Cycotoxic concentr. :
Metabolic activation : with and without
Result : negative
Method : other
Year : 1979
GLP : no
Test substance : as prescribed by 1.1 - 1.4

Remark : Study peer reviewed
Result : No significant colonies appeared all along the streaks.
Source : ATOFINA, Paris-La Défense, France.
Test condition : PRINCIPLE OF THE TEST:
It is a reverse mutation test (like the Ames test).
Two agar layers are poured in a P etri dish. Only the upper layer contains the test substance. The plate is incubated for 2 hours at room temperature in order to let the test substance diffuse in the lower layer, creating thus a gradient of concentrations.
A streaking device, made of 10 sterile micropipets is used to seed the plate. Pipets are dipped in bacterial suspension and drawn down across the plate.
Colonies growing in deep agar are assumed to have been exposed to lower concentration than those growing on the surface.

EXPERIMENTAL CONDITIONS:
- Number of replicates: 3/dose
- Metabolic activation: S9-mix (from male Fischer rats, treated by Aroclor 1254)
- Vehicle: distilled water
- Negative control: no compound
- Positive controls: streptozotocin (S9-) and acetylaminofluorene (S9+)
- Incubation time: 48 hours
- Incubation temperature: 37°C

EXAMINATION:
Number of colonies all along the streaks.

CRITERIA FOR EVALUATING RESULTS:
Negative result: a very pale streak of bacterial growth is seen along the inoculation streak.
Positive result: discrete colonies appear in a pale background lawn with and increased density along the increasing gradient.

Test substance : Source: no data
Batch number: no data
Purity: no data

Reliability : (3) invalid
Concentrations are not checked all along the gradient.
No preliminary study focused on compound diffusion in agar is available.
Tested concentrations are guessed rather than known.

30.12.2002 (42)

Type : DNA damage and repair assay
System of testing : Escherichia coli P3478E
Test concentration : 25 µl/plate (37 µg/plate)
Cycotoxic concentr. :
Metabolic activation : without
Result : negative
Method : other
Year : 1976
GLP : no
Test substance : as prescribed by 1.1 - 1.4

Remark : Study peer reviewed
Result : Diameters of growth inhibition zone for deficient and wild strains were respectively 33 and 30 mm.
Differential is 3 mm and therefore no significant.
Consequently, methane sulfonic acid is not mutagenic.
Source : ATOFINA, Paris-La Défense, France.
Test condition : PRINCIPLE:
Escherichia coli, which are deficient in DNA polymerase and therefore repair-deficient are exposed to test substance. Wild bacteria are simultaneously exposed to the same substance.
Mutagenic effect is measured by the difference of diameters of inhibition growth zone between DNA polymerase deficient and wild strains.

EXPERIMENTAL CONDITIONS:
- Number of replicates: 2
- Vehicle: none
- Negative control: no
- Positive control: dimethyl sulfate, ampicillin and colistin
- Incubation time: 16 hours
- Incubation temperature: 37°C

EXAMINATION:
Zone diameters of DNA polymerase deficient and that of wild strains were measured in millimeters.

measured in millimeters.
Difference was calculated.

CRITERIA FOR EVALUATING RESULTS:
A differential greater than 4 mm was considered as positive.

Test substance : Source: no data
Batch number: no data
Purity: no data

Reliability : (3) invalid
The authors are themselves unforthcoming about the ability of this test to detect real mutagens.

27.12.2002 (43)

5.6 GENETIC TOXICITY 'IN VIVO'

Type : Micronucleus assay
Species : mouse
Sex : male/female
Strain : no data
Route of admin. : gavage
Exposure period : 24h, 48h or 72h
Doses : 0, 20, 100, 500 mg/kg
Result : negative
Method : OECD Guide-line 474 "Genetic Toxicology: Micronucleus Test"
Year :
GLP : yes
Test substance : as prescribed by 1.1 - 1.4

Method : - Equipment: light microscope
- Statistical test: Mann-Whitney U-test

Remark : Study peer reviewed

Result : MORTALITY:
Two males animals dosed with 500 mg/kg were found dead in their cage approximately 21 hours post-dose.
One animal exhibiting pilo-erection, severe rales and loss of facial hair was killed approximately 48h after treatment.
Results from those three animals have been excluded from the analysis

CLINICAL SIGNS:
All surviving male mice of the 500 mg/kg group showed pilo-erection. One of them exhibited rales. One female presented such rales too.
All other animals remained asymptomatic.

NUMBER OF MICRONUCLEATED ERYTHROCYTES PER ANIMALS:
No significant variation in treated animals was reported, whatever the dose after 24, 48h and 72h.
However, chlorambucil group exhibited a significant increase of micronucleated polychromatic erythrocytes.

PROPORTION OF IMMATURE ERYTHROCYTES AMONG TOTAL ERYTHROCYTES (PCE/NCE RATIO):
No real indication of bone marrow toxicity, as evidenced by depression bone marrow proliferation was noted in any group treated by methane sulfonic acid.

The frequency of micronuclei in treated animals was similar to that in concurrent controls. No bone marrow toxicity and no chromosomal damage were evident.

Source : ATOFINA, Paris-La Défense, France.
Test condition : TEST ORGANISMS:

- Source: no data
- Age: no data
- Body weight at study initiation: no data
- Number of animals per dose: 5 males + 5 females

ADMINISTRATION:
- Vehicle: distilled water
- Administration volume: no data
- Frequency of treatment: single administration
- Positive control: chlorambucil (30 mg/kg)
- Negative control: distilled water

EXAMINATIONS:
- Clinical observations
- Mortality
- Tissue examined: bone marrow
- Slide preparation: no data
- Number of cells analyzed per animal: at least 2000

PRELIMINARY STUDY
A preliminary test was first conducted, using dosages of 125, 250, 500, 1000 and 2500 mg/kg.

Test substance : Source: no data
Batch number: no data
Purity: no data

Conclusion : The frequency of micrnuclei in treated animals was similar to that in concurrent controls.
No bone marrow toxicity was evident.

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Risk Assessment, Directive 67/548/EEC

30.12.2002 (44)

5.7 CARCINOGENICITY

5.8.1 TOXICITY TO FERTILITY

5.8.2 DEVELOPMENTAL TOXICITY/TERATOGENICITY

Species : rat
Sex : female
Strain : Sprague-Dawley
Route of admin. : gavage
Exposure period : from gestation day 6 through 15
Frequency of treatm. : once a day
Duration of test : up to gestation day 20
Doses : 25, 100 and 400 mg/ml
Control group : yes, concurrent vehicle
NOAEL maternal tox. : > 400 mg/kg bw
NOAEL teratogen. : > 400 mg/kg bw
Result : No developmental toxicity
Method : OECD Guide-line 414 "Teratogenicity"
Year : 1981
GLP : yes
Test substance : other TS

Remark : Study peer reviewed
Result : - Clinical observations:
No clinical signs that could be attributed to the test article were observed in

No clinical signs that could be attributed to the test article were observed in the treated groups.

- Mortality:
No mortality until day 20 (necropsy)

- Body weight:
Body weight data in the treated groups were not affected by treatment.

- Food consumption:
Food consumption in the treated groups were not affected by treatment.

- Necropsy findings:
Intrauterine growth and survival were unaffected by test article administration at all dose levels.
The foetal malformations observed in the treated groups were considered to be spontaneous in origin.
The developmental variations observed in the treated groups occurred similarly in the control group and/or in a manner which was not dose-related.

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Charles River Breeding Laboratories (Portage, Michigan)
- Age: approximately 70 days old
- Weight at study initiation: 266 ± 9.5 g
- Number of animals: 100 (25 females/dose group)

ADMINISTRATION:
- Vehicle: deionized water
- Concentration in vehicle: 25 or 50 mg/ml
- Total volume administered: 1, 4 or 8 ml/kg

MATING PROCEDURES:
The animals were paired for mating in the home cage of the male.
Positive mating criteria is not precised.

MATERNAL PARAMETERS ASSESSED:
- Clinical observations: yes (twice a day, from day 0 through day 20)
- Mortality: yes (twice a day, from day 0 through day 20)
- Body weight: yes (daily on day 0, day 6 to day 16 and day 20)
- Food consumption: yes (daily on day 0, day 6 to day 16 and day 20)

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
All animals were euthanized on gestation day 20 for a scheduled laparohysterectomy. The uteri and ovaries were examined and the numbers of fetuses, early and late resorptions, total implantations and corpora lutea were recorded. Mean gravid uterine weights and net body weight changes were calculated for each group. The fetuses were weighed, sexed and examined for external, visceral and skeletal malformations and developmental variations.

OTHER EXAMINATIONS: none

Test substance : Source: Elf Atochem
Batch number: C06G
Purity: 70.15%

Conclusion : Based on the results of this study, a dose level of 400 mg/kg/day was considered to be the no observable adverse effect level (NOAEL) for maternal toxicity and developmental toxicity of Methane Sulfonic Acid.

Reliability : (1) valid without restriction

Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

27.12.2002 (45)

Species	:	rat
Sex	:	female
Strain	:	Sprague-Dawley
Route of admin.	:	gavage
Exposure period	:	from gestation day 5 through 15
Frequency of treatm.	:	daily
Duration of test	:	sacrifice on GD20
Doses	:	25, 50, 100, 200 and 300 mg/kg/d
Control group	:	yes, concurrent vehicle
NOAEL maternal tox.	:	50 mg/kg bw
NOAEL teratogen.	:	> 300 ml/kg bw
Result	:	This range-finding study allows to perform a subsequent developmental toxicity study.
Method	:	other: range finding study according to OECD Guide-line 414
Year	:	
GLP	:	yes
Test substance	:	other TS

Remark : Study peer reviewed

Result :

- Clinical observations:
Treatment-related clinical signs observed consisted of rales, labored respiration and gasping in the 100, 200 and 300 mg/kg/d groups. Findings of red material around the nose and/or mouth in the 100, 200 and 300 mg/kg/d groups often correlated with occurences of the aforementioned respiratory abnormalities. These findings appeared to be a function of the dosage concentration; rather the the dosage level, as they were observed with similar frequency in the 100, 200 and 300 mg/kg/d groups, each of which received the test article at a concentration of 50 mg/ml.

- Mortality:
No mortality until day 20 (necropsy)

- Body weight:
slight mean body weight losses and reduction in food consumption occured in the 100, 200 and 300 mg/kg/day groups during gestation days 6-9 when evaluated on a group mean basis. However, several animals in these groups experienced large, transient body weight losses and corresponding large decreases in food consumption on one or more occasions during the first days of the dosing period.
Mean body weights, gravid uterine weights, net body weights and net body weight gains were unaffected by treatment at all dose levels.

- Food consumption:
Food consumption was slightly reduced in the 100, 200 and 300 mg/kg/d groups during gestation 6-9 when the group means were compared with the control group mean. This reduced food consumption seems to be the result of a localized gastro-intestinal effect due to the dosage concentration to these groups (50 mg/ml) and the corrosive nature of the test article.

- Necropsy finding:
No treatment related internal necrops y findings were observed at any dose level.
No effects were observed at any dose level on intrauterine growth and survival. No external developmental variations of malformations were observed in any of the foetuses in the treated groups.

Source : ATOFINA, Paris-La Défense, France.

Test condition : TEST ORGANISMS:
- Source: Cherles River Breeding Laboratories (Portage, Michigan)
- Age: approximately 70 days old
- Weight at study initiation: 261 +/- 10.7g
- Number of animals: 48 (8 females/dose)

- Number of animals: 48 (8 females/dose)

ADMINISTRATION:
- Vehicle: deionized water
- Concentration in vehicle: 25 or 50 mg/ml
- Total volume applied: 1, 2, 4 or 6 ml/kg

MATING PROCEDURES:
The animals were paired for mating in the home cage of the male.
Positive mating criteria in not specified.

PARAMETERS ASSESSED DURING STUDY:
- Body weight gain: on day 0, 6 to 16 and on day 20
- Food consumption: on day 0, 6 to 16 and on day 20
- Clinical observations:
- Mortality: twice a day, from day 0 through day 20
- Examination of uterine content: on day 20, gravid uterine weight, number of corpora lutea, number of implantations
- Examination of fetuses: on day 20, litter size, number of dead fetuses, fetal weight, sex ratio, grossly visible/external abnormalities

Test substance : Source: Elf atochem
Batch number: C06G
Purity: 70.15% in water

Conclusion : Maternal toxicity was observed at dose levels of 100, 200 and 300 mg/kg/d (administered at a concentration of 50 mg/ml), as evidenced by changes in the clinical condition of the animals and inhibition of body weight gain and food consumption.
No maternal toxicity was observed at dose levels of 25 and 50 mg/kg/d (administered at a concentration of 25 mg/ml).
No developmental toxicity was observed at dose levels up to 300 mg/kg/d.

Reliability : (1) valid without restriction

30.12.2002 (46)

5.8.3 TOXICITY TO REPRODUCTION, OTHER STUDIES

5.9 SPECIFIC INVESTIGATIONS

Endpoint	:	other: corrosivity test
Study descr. in chapter	:	
Reference	:	
Type	:	other: in vitro
Species	:	
Sex	:	
Strain	:	
Route of adm in.	:	
No. of animals	:	
Vehicle	:	water
Exposure period	:	4 hour(s)
Frequency of treatm.	:	single exposure
Doses	:	undiluted
Control group	:	yes
Observation period	:	4 hours
Result	:	7min02 (70% methane sulfonic acid) and 3min43 (methane sulfonic acid).
Method	:	
Year	:	
GLP	:	no
Test substance	:	

Remark : study peer reviewed
Result : RESULTS OF PILOT STUDY:
Qualification screen allows CDS use.
Categorization screen allows using the following scoring scale:
Up to 3 minutes: Corrosive (Packing group I)
>3 min but <1h: Corrosive (Packing group II)
>1h but <4h: Corrosive (Packing group III)
>4h: No corrosive

RESULTS OF TEST
See Attached Document
Source : ATOFINA, Paris-La Défense, France.
Test condition : PRINCIPLE OF THE TEST:
The Corrositex assay is used as a standardized and quantitative in vitro corrosivity test. The test is based on the time that is required for the test sample to pass through a biobarrier membrane and produce a change in the Chemical Detection System (CDS).

MATERIAL AND METHOD:
- Biobarrier: membrane disc (unknown composition) placed in a scintillation vial above a chemical detection system.
- Chemical Detection System: unknown composition
- Volume or quantity administered: 500 µl
- Negative control: distilled water
- Positive control: sodium hydroxyde
- number of replicates: 4
- Experiment schedule: Once test substance is placed on membrane disc, vials are continuously observed for the first 10 minutes and then at approximately 5 minutes intervals for up to 4 hours.
- Measured parameter: time is recorded from test substance application until CDS colour change.
- Pilot study: Yes
A preliminary study was conducted in order to determine whether test substance can be detected by CDS or not (qualification screen) and whether the schedule and scoring scale are adapted to the test substance (categorization screen). Methods of such preliminary test are not available.
Test substance : Source: Elf Atochem
Batch number: L06F (methane sulfonic acid), no data concerning 70% methane sulfonic acid
Purity: no data
Attached document : 75-75-2 Corrosivity test.bmp

Test Article		Break Through Time (min:sec)					Packing Group	pH*
Sponsor's Designation	MA Number	Vial 1	Vial 2	Vial 3	Vial 4	Mean		
70% Methane Sulfonic Acid	95BW22	7:20	7:00	6:50	6:57	7:02	II	0
Methane Sulfonic Acid	95BW23	3:50	3:38	3:50	3:35	3:43	II	0
NaOH		12:23	NA	NA	NA	12:23	NA	NA

* 100 µl/ml solution in deionized water
NA - Not Applicable

Conclusion : This in vitro test allows to consider METHANE SULFONIC ACID

(anhydrous and 70% diluted) as corrosive with regard to biomembranes.

Reliability : (2) valid with restrictions
Flag : Material Safety Dataset, Directive 67/548/EEC, Critical study for SIDS endpoint

27.12.2002 (47)

5.10 EXPOSURE EXPERIENCE

5.11 ADDITIONAL REMARKS

(1) Elf Atochem (1987) unpublished study.

(2) Budavari, S. (ed.). The Merck Index - Encyclopedia of Chemicals, Drugs and Biologicals. Rahway, NJ: Merck and Co., Inc., 1989. 938.

(3) Atofina (1999) Material Safety Data Sheet

(4) Atofina (1994) unpublished data

(5) Wiltec research company

(6) LEO, A.J., 1978. Report on the calculation of octanol/water log P values for structures in EPA files. (In ISHOW Database).

(7) Atofina (1987) unpublished study.

(8) Lewis, R.J., Sr (Ed.). Hawley's Condensed Chemical Dictionary. 12th ed. New York, NY: Van Nostrand Rheinhold Co., 1993 751.

(9) Seidell A; Solubilities of Organic Compounds. NY, NY: D Van Norstrand Co. Inc. (1941).

(10) ELF ATOCHEM, Safety Data Sheet, 08/08/1994.

(11) Serjeant ED, Dempsey B; IUPAC Chemical Data Series No. 23. NY,NY: Pergamon Press p. 12 (1979).

(12) Elf Atochem - Acide méthane sulfonique : détermination de la biodégradabilité facile. Essai de disparition du COD - Elf Atochem DCRD Centre d'application de Levallois - Study n°2765/95/A - December 8, 1995.

(13) SNEA(P) - Biodégradabilité ultime de l'acide méthane sulfonique : essai de simulation en maquette de station d'épuration à boues activées aérobies - Atochem DRDI Centre d'application de Levallois - Study n°1228/89/A - October 17, 1989.

(14) Atochem - Biodegradability of methane sulfonic acid - Penwalt - Study n°? - August 23, 1988.

(15) Atochem - BOD and COB of the product MSA - TNO - Study n°R MTS01B - June 12, 1986.

(16) MAGLIETTE RJ, McKINNEY D, VENKAT E (1996) Acclimation and Selection for Methanesulfonic acid (MSA) Degrading Microbial Population using continuous Culture Technique. Bull. Environ. Contam. Toxicol., 57, 406-412.

(17) Microbial metabolism of methanesulfonic acid (Received 12 July 1999/ accepted 10 September 1999). Department of Biological Sciences, University of Warwick. Coventry CV4 7AL. England.

(18) Metabolism of methanesulfonic acid involves a multicomponent monooxygenase enzyme. Department of Biological Sciences and Institute of Education, University of Warwick Coventry CV4 7AL UK. 1996.

(19) Elf Atochem - Methane sulfonic acid: acute toxicity to rainbow trout (Oncorhynchus mykiss) under static conditions - Springborn Laboratories - Study n°97-4-7192 - March 11, 1998.

(20) Elf Atochem - Methane sulfonic acid: acute toxicity to daphnids (Daphnia magna) under static conditions - Springborn Laboratories - Study n°97-12-7191 - March 16, 1998.

(21) ELF ATOCHEM. Determination de l'inhibition de la mobilité de Daphnia magna selon la norme ISO 6341. Centre d'Application de Levallois. Unpublished report, 1989.

(22) OSSI SEPPOVAARA and PERTTI HYNNINEN (1970) On the toxicity of sulphate-mill condensates. Paper och Trea, N°1.

(23) Elf Atochem. Methane sulfonic acid: Acute toxicity to the Freshwater green Alga Selenastrum Capricornutum. Report N°: 97-12-7187, Study N°: 12442.0797.6255.430, October 1997.

(24) Elf Atochem - Acide méthane sulfonique : inhibition de la respiration des boues activées - Elf Atochem DCR Centre d'application de Levallois - Study n°2765/95 - June 19, 1996.

(25) Elf Atochem - Acide méthane sulfonique : détermination de l'inhibition de la croissance des bactéries Pseudomonas putida - Elf Atochem DCRD Centre d'application de Levallois - Study n°95/SAEk/0491/AMP - July 26, 1995.

(26) SCHAFER, E.W. et al, 1983.The acute oral toxicity, repellency and hazard potential of 998 chemicals to one or more species of wild and domestic birds. Arch. Environ. Contam. Toxicol., 12, 355-382.

(27) Elf Atochem - An acute oral toxicity study in rats with methanesulfonique acid, anhydrous - Springborn laboratories - Study n°3255.144 - February 16, 1988.

(28) Elf Atochem - An acute oral toxicity study in rats with 70% methane sulfonic acid - Springborn laboratories - Study n°3255.111 - July 11, 1997.

(29) Union Carbide Corporation, Methane sulfonic acid (MAS) and the potassium salt of MSA, Results of acute peroral intubation and feeding in the diet of rats for 7 days, Chemical Hygiene Fellowship, project report 38-80, June 13, 1975. EPA-OTS0536043.

(30) Eastman Kodak Compagny, Toxicology data on some commercially important conmouns, letter from D.W. Fassett to H.E. Stokinger, dated May 21, 1971.

(31) Pennwalt Corporation - Toxicology report on methane sulfonic acid: skin corrosivity and one-hour inhalation toxicity in mice - September 1, 1976.

(32) Eastman Kodak Company, Toxicology data on some commercially important compounds, letter from D.W. Fassett to H.E. Stokinger, dated May 21, 1971.

(33) Atofina Chemicals (2002) Methane sulfonic acid 70% in water, Acute dermal toxicity in rabbits. MB Research Laboratories report n° MB 02-10135.02.

(34) Pennwalt Corporation, Toxicology report on Methane Sulfonic Acid (Anhydrous), Pharmacology Research, 4/13/78.

(35) Pennwalt Corporation - Toxicology report on methane sulfonic acid: eye irritancy and dermal toxicity - April 13, 1978

(36) Elf Atochem - Delayed contact hypersensitivity study in guinea pigs (Buehler technique) of methane sulfonic acid 70% - Hill Top Biolabs - Study N° 95-9737-21 - October 17, 1995.

(37) Elf Atochem - A 4-week inhalation toxicity study of methane sulfonic acid in albino rats - Wil Research Laboratories - Study n°WIL-160033 - December 17, 1996.

(38) Elf Atochem - A 5-day inhalation toxicity dose range-finding study of methane sulfonic acid in rats - Wil Research Laboratories - Study n°WIL-160032 - January 22, 1996.

(39) SNEAP (Société Nationale Elf Aquitaine Production) - METHANE SULFONIC ACID : Ames test (Reverse mutation assay on Salmonella typhimurium) - Sanofi Recherche - Study n°CEL473A/473B - 10 Mai 1990

(40) Pennwalt Corporation, Methane Sulfonic Acid : Assessment of mutagenic potential in histidine auxotrops of Salmonella typhimurium, Life Science Research, report n° 89/PSV035/0286, August 23, 1989.

(41) Zeiger E and Pagano DA. Mutagenicity of the human carcinogen treosulphan in Salmonella. Environmental and Molecular Mutagenesis 1989 ; 13 : 343-346

(42) McMahon Re, Cline JC and Thompson CZ. Assay of 855 test chemical in ten tester strains using a new modification of the Ames test for bacterial mutagens. Cancer Res . 1979 ; 39 : 682-693

(43) Fluck, E.R., Poirier, L.AL and Ruelius, H.W. (1976) Evaluation of a DNA polymerase-deficient mutant of E. coli for the rapid detection of carcinogens, Chem.-Biol. Interact., 15, 219-231.

(44) Pennwalt Corporation, Methanesulfonic acid: Assessment of clastogenic action on bone marrow erythrocytes in the micronucleus test, Life Science Research, report n° 89/PSV036/0243, May 26, 1989.

(45) Elf Atochem - A developmental toxicity study of methane sulfonic acid in rats - Wil Research Laboratories - Study n°WIL-160048 - December 18, 1996.

(46) Elf Atochem - A dose range-finding developmental toxicity of methanesulfonic acid in rats - WIL Research Laboratories - Study n°WIL-160047 - spetember 4, 1996.

(47) Elf Atochem - Corrositex continuous time monitor assay: 70% methane sulfonic acid and methane sulfonic acid - Microbiological Associates - Study n°A95BW22-A95BW23.520001 - December 7, 1995.